Canad. Med. Ass. J.
July 14, 1962, vol. 87

Incidence of Adenovirus Infection:

A Family Study

J. JONCAS, M.D., A. MOISAN, M.D.* and V. PAVILANIS, M.D., *Montreal*

THE incidence of adenovirus infection has now been surveyed in almost all parts of the world by serological studies with or without isolation of viruses.[3, 11, 13, 20-23, 31-33] Most of these surveys have been made in relatively isolated populations such as military camps,[1, 12, 26, 29] summer camps[25] for children, or children or adults in other closed communities.[4]

A few surveys, however, have been done on civilians[23, 30, 33] at random or in a family population. The best-known family population study is the one made in Cleveland, Ohio.[16-18] The incidence of adenovirus infection in this latter study was found to be very low; only 1 to 5% of surveyed persons in that group showed a significant rise in antibody to any one of Adenoviruses types 1 to 7 over a period of 10 months.

The results from a previous study on cases of diarrhea suggested a much higher incidence of these infections among infants and children in Montreal.[15] For this reason, it was decided to conduct a large-scale investigation of the incidence of adenovirus infection in family groups.

STUDY GROUP

Forty-six families totalling 223 members were available for the study (Table I); they were all from the southwestern section of Montreal and were of definitely lower income than the average.

Members of these families were attending two of the public health clinics in the area, for regular check-ups and routine immunization. A visiting nurse was available to help in the study. A pediatrician (J.J.) was notified whenever an illness was reported; the parents were contacted by telephone, and in most cases the child was seen either at home or on appointment at the clinic within the next few days. A record of each family was kept by the pediatrician.

Each of the families was first seen in the clinic in the fall of 1959 over a period of two months. A specimen of blood was taken and the purpose of the study was explained to them, as was the technique of collecting throat swabs or washings and rectal swabs. This method was adopted to avoid delay in taking specimens, thereby enhancing the chance of virus recovery. Whenever possible, however, specimens were taken either by the visiting nurse or the pediatrician.

TABLE I.—THE POPULATION UNDER STUDY

Age (in years)	*Number of persons*	%
Children: 0- 1	16	7.1
1	17	7.6
2	22	9.8
3	22	9.8
4	20	8.9
5	21	9.4
6	12	5.3
7	12	5.3
8	9	4.0
9	7	3.1
10-14	9	4.0
Adults	56	25.1
Total	223	100.0

Parents were told to report any respiratory, enteric or febrile illnesses as well as any combination of these; particular emphasis was placed on the importance of reporting conjunctivitis. Contact by telephone was renewed with each family at least twice during the period of study.

A second specimen of blood was taken at the end of the study in the spring of 1960; the average period between the two samples was four to six months.

METHODS

The throat and rectal swab specimens were placed immediately in the freezer compartment of an ordinary refrigerator; they were picked up within a few days and were kept at a temperature of minus 30° C. while being brought to the laboratory. Assay was by inoculation of each specimen into at least three tubes of HeLa cell cultures;[15] the specimens were stored for a period of up to one year, following collection. The specimens were observed on inoculated tissue culture for an average period of 40 to 45 days.

Complement fixation and neutralization tests for antibodies to Adenovirus types 1 to 7 were done on 223 paired sera. Unpaired sera which were available from several other members of the families in the study and from families which dropped out of the study were not included in the survey.

A screening operation was first done with a serum dilution of 1:8. Then, serial dilutions of the paired sera were made as follows: 1/16, 1/64, 1/256 and 1/1024. A rise in antibody level was considered significant either if a fourfold or greater increase occurred or a conversion from negative at 1/8 dilution to positive at or beyond 1/16 was demonstrated.

From the Institute of Microbiology and Hygiene of the University of Montreal.
*Present address: Hôpital St-Sacrement, Québec, Qué.
This work was partially aided by a grant from the Ministry of Health of the Province of Quebec (Federal-Provincial Public Health Grant).

TABLE II.—CORRELATION OF EPISODES OF ILLNESS WITH VIRUSES ISOLATED

Name	Age in years	Adenovirus antibodies: Complement fixation, First and second specimens, Collection dates	Titre	Neutralizing: 1	2	5	6	7	Clinical symptoms	Virus isolated
S.L.	25	Sept. 10/59	0- 32	8- 8	256-256	16- 8	0- 0	0- 0		
R.L.	4	Feb. 18/60	0- 0	64- 64	256- 32	0- 0	12- 16	32- 32		
J.L.	1		0-256	0-180	152-2000	0- 0	0- 0	0- 0	Feb. 3-7/60 J.L. Febrile U.R.I. with cough	
M.L.	<1 year		0- 0	0- 0	0-600	0- 0	0- 0	0- 0	Feb. 16-20/60 M.L. Febrile U.R.I. with L. conjunctivitis	T.* Feb. 18/60 Adeno 2
E.G.	32	Oct. 8/59	4- 0	96-256	64- 64	0- 0	0- 12	8- 8	Oct. 6-12/59	
Je.G.	9	Jan. 12/60	64- 0	64-128	128- 96	0- 0	0- 16	256-256	E.G., Ma.G., M.G. Febrile rhinopharyngitis with cough.	R.** Oct. 9/59 Cox. B5
Jo.G.	8		256-128	256-256	128-128	0- 0	0- 0	32- 16		
Ja.G.	7		64- 0	32- 16	64- 32	0- 0	0- 0	0- 0	Ja.G. Febrile gastroenteritis Oct. 10/60	T. Oct. 9/59 Adeno 1
Ma.G.	6		256- 32	90- 90	250-250	0- 0	0- 0	16-180	Nov. 5-12/59	
M.G.	3		0- 0	64- 64	0- 0	0- 0	0- 0	16- 90	Ma.G., M.G. Febrile U.R.I. with cough	R. Nov. 5/59 Adeno 7 R. Nov. 9/59 Adeno 7
									March 14-24/60 E.G., Jo.G. Febrile rhinopharyngitis Ja.G. Diarrhea and vomiting—one day	R. March 18/60 Polio 1
									Ma.G., M.G. Afebrile rhinopharyngitis	R. March 16/60 Polio 1
My.K.	23	Oct. 6/59	0-128	128-256	0- 12	0- 0	32- 32	0- 0	Nov. 24-30/59	
R.K.	6	Mar. 29/60	0-128	256-128	16- 8	0- 0	8- 8	0- 0	R.K., Me.K., H.K., C.K. Febrile U.R.I. with cough	
Me.K.	5		0- 0	64-128	16- 8	0- 0	0-128	0- 0		
H.K.	4		0- 0	64- 64	8- 8	0- 0	8- 8	0- 0		
C.K.	2		16- 0	512-1024	16- 16	0- 0	0- 12	0- 0	March 12-20/59	
P.K.	<1 year		N.D.-N.D.						P.K. Spasmodic bronchitis March 27-30/59 My.K. Afebrile pharyngitis H.K., C.K. Febrile U.R.I. with cough	R. March 17/60 Adeno 1
J.B.	29	Oct. 15/59	0- 16	64- 32	8- 8	0- 0	32- 32	0- 0	Nov. 5-12/59	
Ga.B.	24	Mar. 31/60	0- 0	0- 0	32- 16	32- 32	0- 0	0- 0	Ga.B., A.B., J.B., Ge.B. Afebrile rhinitis	
A.B.	5		64-128	0- 0	32- 32	0- 32	0- 0	0- 0		
J.B.	4		64- 64	0- 0	32- 16	32- 12	0- 0	8- 8		
Ge.B.	2		128- 8	0- 0	16- 12	8- 8	0- 0	0- 0		
Ma.B.	1		0- 64	0- 0	0- 64	128-150	0- 0	0- 0	Ma.B. Measles-like picture: rash 36 hours, no Koplik spots Jan. 21-28/60 Ge.B. Afebrile rhinitis with cough Ma.B. Afebrile rhinitis with cough, bilateral conjunctivitis March 1-4/60 Whole family: Fever and vomiting March 20-30/60 Ga.B. Afebrile pharyngitis Ma.B. Afebrile rhinitis with cough	T. Jan. 25/60 Adeno 2
T.A.	28	Oct. 27/59	32- 64	128-512	0- 0	128-512	0- 0	0- 0	Jan. 12-21/60	
Ra.A.	7	April 5/60	0- 0	32-128	32-128	0- 0	0- 0	0- 0	Ro.A. Afebrile rhinitis H.A. Febrile rhinopharyngitis	
Ro.A.	5		0- 32	64-256	32- 32	32-32	0- 0	0- 0	J.R.A. Febrile rhinitis with L. conjunctivitis	
H.A.	4		0-128	32-128	512-512	0-128	0- 0	0- 0	March 14-20/60 J.R.A. Febrile rhinobronchitis	T. March 18/60 Adeno 1
J.R.A.	1		0- 32	0- 90	0- 0	0- 0	0- 0	0- 0	April 3-7/60 T.A., Ro.A., J.R.A. Afebrile rhinitis	
R.M.	32	Oct. 29/59	0- 0	512-128	32- 32	8- 8	32- 32	0- 0	Jan. 16-26/60	
		April 21/60							D.M. Febrile respiratory enteric syndrome	
L.M..	11		0- 64	0- 0	128-512	128-128	0- 0	0- 0	Jan. 27- Feb.2/60, W.M., C.M. Febrile rhinitis with cough	T. Jan. 27/60 Adeno 2
A.M.	9		0-128	0- 0	128-512	0- 64	8- 8	0- 0		
K.M.	8		0- 64	0- 0	512-128	32- 32	0- 0	0- 0		
C.M.	2		64-128	0- 0	128-256	0- 0	8- 8	0- 0		
W.M.	1		0- 0	0- 0	0- 16	0- 0	0- 0	0- 0		
D.M.	<1 year		N.D.-N.D.							

*T = throat swab.
**R = rectal swab.

RESULTS

The first specimens of sera were collected from September 3, 1959, to November 10, 1959 and the second specimens from February 2, 1960, to May 12, 1960. In one exceptional family where six viruses were isolated, the second serum specimens were taken earlier, on January 12, 1960.

During this period a total of 242 illnesses was reported: 46 among adults and 196 in children. Most of these were respiratory infections, either afebrile or febrile.

The most frequent symptoms were rhinitis and cough. Cough was reported 85 times, with fever three times in adults and 35 times in children; the

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TABLE III.—CONJUNCTIVITIS IN FAMILIES WHERE NO ADENOVIRUS WAS ISOLATED

Name	Age in years	*Adenovirus antibodies*							Clinical symptoms	Virus isolated
		Complement fixation		*Neutralizing*						
		First and second specimens Collection dates	Titre	1	2	5	6	7		
P.R.	33	Oct. 8/59 Mar. 31/60	0- 0	64-128	32- 32	8- 8	0- 8	8- 8	Oct. 15-28/59 G.R. Biphasic febrile U.R.I. with vomiting	R.* Oct. 14/59 Polio 1
J.R.	9		0- 0	256-256	128-128	0- 0	0- 0	0- 0	Dec. 1-21/59 P.R. Isolated L. conjunctivitis	
Mi.R.	5		16- 0	8- 8	8- 8	0- 0	0- 32	8- 8	Dec. 1-7/59 J.R., Mi.R., Ma.R. } Afebrile rhinopharyngitis	
Ma.R.	4		0- 32	64- 64	8- 16	0- 0	8- 16	0- 0		
G.R.	1		32- 0	0- 0	0- 0	0- 0	0- 0	0- 8	G.R. Febrile rhinopharyngitis	
H.D.	39	Oct. 8/59 Mar. 29/60	8- 0	8- 8	0- 0	0- 0	0- 32	0- 0	Oct. 1-8/59 H.D., N.D., J.D. } Afebrile rhinopharyngitis	
N.D.	7		32-128	8- 32	8- 8	0- 0	32- 32	8- 8		
J.D.	5		0- 0	1024-1024	0- 0	0- 0	64- 32	128- 32		
D.D.	1		0- 0	0- 0	0- 0	0- 0	0- 0	0- 0	Dec. 23— Jan. 7/60 Family: Afebrile U.R.I. with cough Feb. 1-9/60 H.D. Rhinitis with L. conjunctivitis N.D., J.D. } Rhinitis with cough D.D. Rhinitis with bil. conjunctivitis March 23-29/60 Family: Afebrile rhinolaryngitis	
D.B.	31	Oct. 22/59 April 7/60	0- 0	32- 32	1024-1024	0- 0	0- 0	0- 0	Nov. 1-8/59 Do.B. Febrile U.R.I.	
A.B.	29		0- 0	128-128	32- 32	0- 0	8- 8	0- 0	C.B. Febrile U.R.I. with conjunctivitis	
L.B.	7		0- 0	128-128	128-128	128- 64	8- 8	0- 0	Dec. 5-12/59 D.B., A.B., Do.B. } Afebrile U.R.I. with cough	
Do.B.	6		0- 64	128-128	512-512	32- 32	32- 32	0- 0		
C.B.	5		32-128	1024-1024	512-512	0- 0	0- 12	0- 0		
M.B.	4		32- 0	32-128	128-128	0- 0	16- 32	0- 0		
G.P.	32	Nov. 5/59 April 21/60	0- 0	512-512	512-512	32- 32	0- 0	0- 0	Nov. 1-6/59 Ma.P. U.R.I. + otitis + conjunctivitis	
M.P.	28		0- 16	256-128	256- 64	0- 0	32- 32	0- 0	J.P. U.R.I. + conjunctivitis	
L.P.	5		0- 64	128-128	128-128	8- 8	0- 0	0- 0	P.P. U.R.I. + otitis + conjunctivitis	
D.P.	4		0- 0	128-128	512-512	0- 0	0- 0	0- 0	Nov. 25/59 P.P. Varicella	
Ma.P.	3		0- 0	1024-1024	128-512	0- 0	0- 12	0- 0	Jan. 12-20/59 Family: U.R.I. with cough	
J.P.	3		0- 32	512-512	512-512	32-256	0- 32	0- 0	April 15-21/60 P.P. Afebrile rhinitis	
P.P.	1		0-256	512-128	128-128	0- 0	0- 0	0- 0		

*R = rectal swab.

physical signs diagnostic of bronchitis were recorded in 11 of these cases, 13%. Rhinitis was reported 90 times, but fever was associated with this symptom only 16 times and only in children. Sore throat or the physical sign of pharyngitis associated with this symptom was recorded 35 times; fever was present in 16 of these patients, four adults and 12 children. Muscle pains and significant lymphadenopathy occurred infrequently.

Conjunctivitis, isolated (one case) or combined with other respiratory symptoms, was recorded 10 times. In two cases an Adenovirus type 2 was isolated, and a fourfold rise in antibodies was shown (Table II). In most of the other cases a satisfactory correlation with adenovirus infection could be established with the help of serologic findings alone (Tables II and III): seven cases with Adenovirus type 6 and one case with Adenovirus type 5.

The occurrence of high fever without other symptoms or signs was reported in 18 cases. In one family, out of six children who had this symptom at the same time, herpes simplex virus was isolated from the stools of two. The results of the neutralization test appear in Table IV. In view of the already high level of neutralizing antibodies in the first serum specimen taken one month prior to the disease, the interpretation of these findings is difficult.

Diarrhea was reported 16 times; it was accompanied by fever in seven cases. In only three could a presumptive diagnosis of adenovirus infection be entertained. Two of these cases appear in Table II (families G. and M.). An additional patient, aged one year, showed a rise from 0 to 1/670 in neutralizing antibodies to Adenovirus type 1, but no antibodies to the other types and no other diseases reported during the period of study. All three syndromes were febrile and two were labelled as respiratory-enteric syndrome.

During the period of the study, 456 specimens were collected from 228 subjects in the form of rectal or throat swabs taken at the time of an illness or occasionally at random. The discrepancy between illnesses reported and swabs collected is mainly accounted for by a number of illnesses reported at the time of the periodic contact by telephone, a time which was too late for isolation studies.

TABLE IV.—HERPES VIRUS INFECTION

Age	Symptoms and signs	Virus isolation		Herpes neutralizing antibodies			
		Date		Date	Titre	Date	Titre
2 yrs.	Fever of unknown origin—4 days	23-11-59	Herpes s. (R)	29-10-59	90	28-4-60	600
2 yrs.	Fever of unknown origin—4 days	23-11-59	Herpes s. (R)	29-10-59	45	28-4-60	350

R = rectal swab.

From these 456 specimens, 21 viruses were isolated, eight from a throat swab and 13 from a rectal swab. There were 10 Poliovirus type 1 isolations, three from throat swabs and seven from rectal swabs. Two herpes simplex viruses were isolated from rectal swabs, and one Coxsackie B5 virus from a rectal swab in an adult. Eight adenoviruses were isolated, five times from a throat swab and three from a rectal swab. The presence of the adenoviruses could be satisfactorily correlated with the illnesses reported (see Table II); except in one patient, a fourfold rise was demonstrated in all paired sera. The correlation, however, between the enteroviruses isolated and the diseases reported presented a problem which will be discussed later.

The complement fixation test was done on paired sera in 221 cases. In view of the fact that the presence of these antibodies, as shown by other workers[2] as well as in our own laboratory, is better correlated with a recent infection and has a tendency to drop relatively fast within as short a period as the length of this study, the results of these tests for the first and second sera will be given separately. Of the 221 first sera, 81 (36.6%) were positive at a dilution of 1/8 or higher and 44 (20%) at a dilution of 1/64 or higher. A titre of 1/64 or higher was found in 24 of the 46 families, and a level of 1/128 or higher in 12 families.

Of the second sera, 120 were positive at a dilution of 1/8 or higher (54.3%), and 81 were positive

TABLE V.—INCIDENCE OF ADENOVIRUS NEUTRALIZING ANTIBODIES IN THE 223 MEMBERS OF 46 MONTREAL FAMILIES

A: PRESENCE OF ANTIBODIES IN 1/8 DILUTION: SPRING 1960

Type of virus	0-4 yr. (97)*	5-9 yr. (61)	10-14 yr. (9)	Adults (56)	Total (223)
1	63.9%	67.2%	66.6%	83.9%	70.0%
2	55.6%	78.7%	88.8%	85.7%	70.8%
3	12.3%	19.6%	0%	35.7%	19.7%
4	0%	0%	0%	7.1%	1.7%
5	26.8%	50.0%	22.0%	64.2%	42.1%
6	24.7%	36.0%	55.5%	50.0%	35.8%
7	8.2%	21.3%	0%	17.8%	13.9%

B: FREQUENCY OF A FOURFOLD INCREASE IN ANTIBODY TITRE: BETWEEN FALL 1959 AND SPRING 1960

Type of virus	0-4 yr. (97)*	5-9 yr. (61)	10-14 yr. (9)	Adults (56)	Total (223)
1	15.3%	11.4%	0%	5.2%	11.0%
2	12.3%	10.0%	22%	7.1%	10.7%
3	2.0%	0%	0%	1.8%	1.3%
4	0%	0%	0%	1.7%	0.44%
5	4.1%	6.5%	0%	3.5%	4.4%
6	4.1%	6.5%	11%	10.7%	6.7%
7	1%	3.2%	0%	0%	1.3%

*Figure in parentheses = No. of persons.

TABLE VI.—RELATION OF THE PRESENCE OF POLIOVIRUS TO SYMPTOMS OBSERVED

Age (years)	Symptoms and signs	Virus isolation		Homotypic polio neutralizing antibodies			
		Date		Date	Titre	Date	Titre
2	Fever of unknown origin—2 days	9- 9-59	Polio 1 (T)	3- 9-59	32	22- 2-60	ND*
1	Febrile U.R.I. with cough	6-10-59	Polio 1 (T)	10- 9-59	0		ND
7	Diarrhea and vomiting	18- 3-60	Polio 1 (R)	8-10-59	0	12- 1-60	512
6	Afebrile rhinopharyngitis	16- 3-60	Polio 1 (R)	8-10-59	0	12- 1-60	512
5	Inapparent infection	14-10-59	Polio 1 (T)	6-10-59	512	31- 3-60	512
4	Inapparent infection	14-10-59	Polio 1 (R)	6-10-59	512	31- 3-60	512
6 mo.	Inapparent infection	16-10-59	Polio 1 (R)	6-10-59	512	31- 3-60	128
1	Biphasic febrile U.R.I. with vomiting	14-10-59	Polio 1 (R)	8-10-59	0	31- 3-60	512
2	Febrile pharyngitis with sore neck (whole family)	15-12-59	Polio 1 (R)	20-10-59	512	7- 4-60	512
8	Inapparent infection	25-11-59	Polio 1 (R)	3-11-59	0	7- 4-60	0

*ND = not done. T = throat swab. R = rectal swab.

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at a dilution of 1/64 or higher (36.6%). Thirty-three families showed antibody at a level of 1/64 or higher, and 29 had levels of 1/128 or higher.

From these findings alone the existence can already be anticipated of a very high proportion of adenovirus infection in our study population, and especially during the study period.

The percentage of fourfold increases in antibody titre per individuals and per families is in keeping with this; 81 (36.6%) of the individuals (19 adults and 62 children) in 36 of the 46 families in this study showed a fourfold rise. The correlation between these increases in antibody titre, the illnesses reported and the results of the neutralization tests will be outlined subsequently.

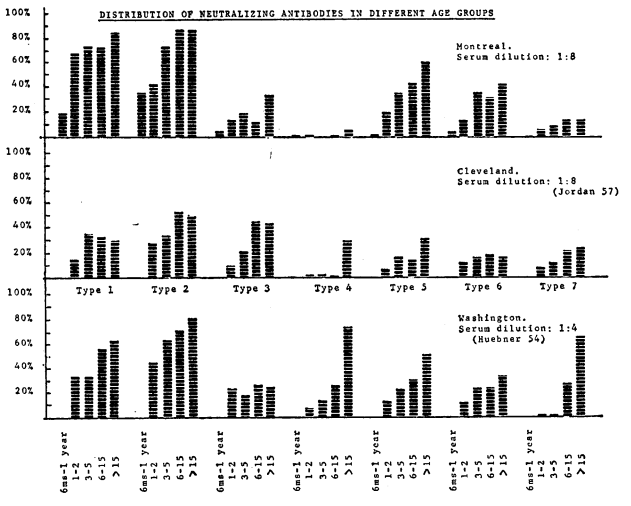

Fig. 1

The results of the neutralization tests are summarized in Table V. If these are compared with the findings of other workers[13, 18, 23] (Fig. 1), a marked difference is found in both the incidence of antibodies to the so-called "sporadic types of adenoviruses"—namely types 1, 2 and 5—and in the frequency of infection or reinfection with these agents in Montreal. Usually all of the members of one family have antibodies against the same types of adenoviruses or they all have no antibodies (Table II). This indicates that adenovirus infections, at least those due to the sporadic types, are family infections and may be compared to those resulting from the enteroviruses.

Conversely, the incidence of antibodies to Adenoviruses types 3, 4 and 7 was comparatively low in our population at the time of this study, and the percentage of infection with these agents was also low. The incidence of Adenovirus type 4 antibody is almost negligible. Both the high incidence of Adenovirus type 6 antibodies and the high frequency of infection with this agent as indicated by fourfold antibody increase are worthy of note.

COMMENTS

The correlation of virus with disease will be attempted only in those patients from whom an adenovirus was isolated; this is done in the hope that the accumulation of such data[8, 9, 14, 24, 27] as these will lead to a more exact description of the spectrum of symptoms likely to follow an infection with Adenoviruses types 1, 2, 5 and 6.

The clinical picture in the family contacts at the time of the isolation of a virus in a particular case is recorded in Table II. For a complete epidemiological picture, all of the illnesses reported during the period of study have been recorded. As mentioned before, the correlation between adenoviruses isolated, neutralizing antibodies detected and clinical disease observed is relatively good. Such a correlation is probably attained only if the virus is isolated from the throat or conjunctiva; furthermore, the younger the child is, the closer the correlation appears to be. This is probably because previous contact with any member of that group of viruses will lead to a heterotypic response in neutralizing antibodies[7, 10, 34] as well as in other immune mechanisms which will afford some protection to the host. This heterotypic response apparently alters the clinical disease in a subsequent infection, even to the point of inapparent infection. These phenomena of recall response and of broadening of antibody spectra with successive infections have recently been stressed by Henle *et al.*[10] Similarly, whether or not a high rate of endemic infection to Adenovirus types 1, 2, 5 or 6 afforded protection here against the epidemic types is open to speculation. But this would provide an explanation for our findings of a low incidence of antibodies to types 3, 4 and 7 and a low rate of infection to these types of adenoviruses.

Family A. in Table II is an illustration of the results of a primary infection with Adenovirus type 1 in baby J.R.A.; the other members of the family experienced a reinfection, as evidenced by a fourfold rise in antibodies without clinical disease. Despite the failure to isolate Adenovirus type 5 in this family, it was assumed that the disease reported in January, upper respiratory infection with conjunctivitis, was the result of a type 5 infection; again the mother (T.A.) shows a fourfold rise in neutralizing antibodies to Adenovirus type 5 without disease. On the other hand, patient A.B. in family B. and patient A.M. in family M. show a diagnostic rise in antibodies to Adenovirus 5 in the absence of significant disease. Finally, C.M. and Ge.B. afford examples of disease presumably due to a reinfection with Adenovirus 2 without a fourfold rise in antibodies. Therefore it seems obvious that inapparent infections as well as reinfections with or without overt disease exist. Furthermore, reinfection with clinically apparent disease may occur either with or without a fourfold rise in neutralizing antibodies (M.G. and Ma.G.), illustrating the obscure nature of immune mechan-

isms in viral infections. Consequently the correlation of the clinical illness with the rest of our data in those families in whom virus isolation was not obtained is practically impossible. However, it seems worthy of note that in these families a high rate of respiratory diseases with cough and rhinopharyngitis as leading symptoms occurs in parallel with an unusually high frequency of adenovirus infections, 36.6% by both neutralization and complement fixation tests.

During the study period, six viruses were isolated from different members of family G. (Table II) at the time of illness; clinically there were only three such episodes involving four, two and six members of the family, respectively. Except for the second episode, we were unable to assign an etiological relationship to any of the agents isolated.

The first disease reported on October 9, 1959, at the time of the first serum sampling, involved the mother and three children; E.G. and Ma.G. had similar illnesses which included muscle aches in addition to a rhinopharyngitis; a Coxsackie B5 was isolated from the mother (E.G.), but no antibodies could be found in either of the two sera. On the other hand, Ja.G. had fever, diarrhea and vomiting for one day, on October 10, 1959, and M.G. had a febrile rhinopharyngitis. As this last child was only three years old, it was quite difficult to ascertain whether he had muscle pains or not; Adenovirus type I was isolated from his throat. The neutralizing antibody level in the first serum specimen from this child taken during the illness was 1/64 and remained at that level in the second serum while the antibody titre in the mother rose somewhat. Although the etiological agent in this episode is unknown, we suspect the adenovirus because persistence of enteroviruses in the stools without active infection or disease seems established, whereas persistence of adenoviruses in the throat without evidence of clinical infection is doubtful.[5, 28]

The second episode which involved only Ma.G. and M.G., both of whom excreted Adenovirus type 7 in their stools, was interpreted as a reinfection with this virus because of an increase of more than fourfold in neutralizing antibodies from a level of 1/16 in both cases. However, in view of reports[6] of persistent excretion of adenoviruses in stools and the level of 1/16 one month prior to the disease, this interpretation is subject to argument.

The third illness, which was reported in March 1960, involved the parents and four of the five children, and presented itself as an acute rhinopharyngitis of 10 days' duration; the illness was febrile in the parents and in Jo.G. and afebrile in the others. Ja.G. responded to this infection in the same manner as he had responded to a previous one (he had diarrhea and vomiting of one day's duration on March 18, 1960). Poliovirus type 1 was isolated from the stools of Ja.G. and Ma.G. However, the neutralizing antibodies to Poliovirus 1 rose from 0 in the first serum to over 1/512 in the second; the latter was obtained on January 12, 1960, in both cases. This seroconversion occurred two months before the last clinical episode during which Poliovirus type 1 was isolated. Despite the virus isolation, the study of antibodies indicates that this last episode was not related to poliovirus infection. The causal agent, if viral, possibly could have been isolated if the poliovirus had not destroyed the culture so rapidly. These findings suggest a third etiological possibility with respect to the first disease reported in October 1959.

This is a good example of the difficulties experienced in trying to correlate the 10 polioviruses isolated in this study with clinical disease recorded in these families (Table VI). If the results of the neutralization tests are taken into account and it is assumed that a poliovirus isolated from the throat has more significance than that isolated from the stools, an acceptable correlation with coincident disease is established in only three out of the 10 cases. These limited findings are tentatively interpreted as evidence that clear-cut disease due to poliovirus ends in October and that the survival of the virus until the next season is made possible by the presence of asymptomatic carriers in the population throughout the winter.

The correlation between the increase of antibodies in the neutralization and complement fixation tests is poor, especially if individual values are taken instead of family values. This low degree of correlation is disturbing, but several explanations of it are available: Infants in this study, with only one exception, did not develop complement fixation antibodies in response to infections, but did develop neutralizing antibodies (example, M.L. in Table II). On detailed analysis it becomes obvious that the correlation is better for Adenoviruses types 2 and 1; the fact that our antigen for the complement fixation test is prepared from Adenovirus type 2 may explain this discrepancy. It has been suggested[19] that the correlation between the two tests is better when this supposedly group-specific antigen is prepared from the homologous type of virus, indicating a variable specificity of this antigen depending on its source. Finally, the existence of types of Adenovirus other than 1 to 7 for which neutralizing antibodies were not determined provides another explanation for the discrepancy noted between these two tests. That the number of adenovirus infections proved by a fourfold rise in antibodies by the complement fixation test is exactly the same as the one found by the neutralization test (81 or 36.6%) is purely coincidental. Nevertheless, the fact that the values for tests are in the same range supports the conclusion that there is a high incidence of adenovirus infection in the population under study.

The technique of sample collection and the delay in tissue culture inoculation may account for the low rate of adenovirus recovery. These adverse

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conditions probably result in a more frequent recovery of the virus when a large specimen is taken. In this study, a virus could be isolated only in the younger age group where the growth of the virus was unimpeded by fully developed immune mechanisms and allowed a large yield from the sampling sites. The rise of homotypic antibodies in siblings (often fourfold) (Table II) supports the contention that an infection or a reinfection, with or without apparent disease, actually occurred despite the failure to isolate the virus from the affected individuals.

SUMMARY

A high rate (36.6%) of adenovirus infection has been demonstrated in 46 Montreal families by means of complement fixation and neutralization tests on 223 paired sera over a period of six months. These infections were mainly of the so-called sporadic types, namely 1, 2, 5 and 6. Such a high incidence is probably in part the result of the low standards of living of the families under study.

A high incidence of antibodies to Adenovirus types 1, 2, 5 and 6 is also shown as opposed to both a low incidence of antibodies to types 3, 4 and 7 and a low rate of infection with these types of adenoviruses; a tentative explanation for these findings is offered.

A total of 21 viruses was isolated, eight from a throat swab and 13 from a rectal swab; there were 10 polioviruses, eight adenoviruses, two herpes simplex viruses and one Coxsackie B5 virus. In general the presence of the adenoviruses could be satisfactorily correlated with the illnesses reported but not that of the other viruses.

A correlation between the diseases reported during the study period, the viruses isolated and the results of the neutralization and complement fixation tests is attempted, and the many pitfalls encountered in such a process are outlined.

The authors wish to acknowledge the valuable co-operation of the late Dr. B. Benjamin, of Dr. D. S. Fleming and of the nursing staff of the Child Health Association, without which this work would not have been possible.

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RÉSUMÉ

L'incidence des infections à adénovirus parmi les membres de 46 familles de Montréal fut déterminée par les épreuves de neutralisation et de fixation du complément faites sur 223 paires de sérum à 6 mois d'intervalle. Le taux de ces infections telles que démontrées par l'augmentation du titre des anticorps par une marge d'au moins 4 dilutions s'est avéré très élevé (36.6%). Ces infections cependant furent surtout du type sporadique tel que 1, 2, 5 et 6. Une incidence aussi élevée relativement aux déterminations faites ailleurs est peut-être en partie le reflet du niveau de vie économiquement inférieur des familles mises à notre disposition.

Le nombre de sujets possédant des anticorps à dilution 1:8 ou plus aux types 1, 2, 5 et 6 fut très élevé alors qu'aux types 3, 4 et 7 ce nombre fut relativement bas; un peu paradoxalement ceci ne sembla pas augmenter les chances d'infections à ces derniers types.

Au total, vingt et un virus furent isolés durant la période étudiée, 8 provenant d'un prélèvement oropharyngé et 13 d'un prélèvement rectal. Le décompte s'établit ainsi: 10 virus polio, 8 adénovirus, 2 virus herpès simplex, et 1 Coxsackie B5. La corrélation entre adénovirus isolés, données sérologiques et maladies déclarées put être établie avec satisfaction, en règle générale. Cependant dans le cas de quelques adénovirus et pour la plupart des autres virus, une telle tentative mit en lumière une foule d'embûches rendant l'interprétation des résultats beaucoup plus difficile.

PAGES OUT OF THE PAST: FROM THE JOURNAL OF FIFTY YEARS AGO

A strong feeling exists in the popular mind that bromides in epilepsy are not only useless, but actually harmful. Before 1857, the date of the introduction of bromides, many cures were reported by the users of such drugs as zinc oxide, silver nitrate, and belladonna. Modern statistics differ from these very little. Writers such as Pierce Clarke argue that hence bromides are neither necessary nor desirable.

The actual effect of bromides in epilepsy is variable and uncertain. In perhaps twenty-five per cent. of cases the attacks are either temporarily or permanently arrested. This class probably includes the spontaneously curable cases of the disease and here bromides are desirable. In a second group amounting to a further twenty-five per cent. the fits become less frequent. This may be looked on as the common, temporary result of bromide treatment. In a third group, composed of fifty per cent. of all cases, bromides either have no effect or else an absolutely deleterious one. Certainly all cases of recent origin should be given the benefit of the remedy for a time, although, in addition, occupation and general physical and mental hygiene should be attended to.—The Ontario Medical Association: *Canad. Med. Ass. J.*, 2: 604, 1912.