SPORADIC ACUTE RESPIRATORY INFECTIONS IN ADULTS WITH SPECIAL REFERENCE TO ADENOVIRUS INFECTIONS

By SYBILLE STOVIN

Bacteriology Department of the Royal Free Hospital School of Medicine, Royal Free Hospital, Gray's Inn Road, London, W.C. 1

INTRODUCTION

Certain types of the adenoviruses are now established as the causative agents of a significant proportion of acute respiratory disease (ARD) and virus pneumonia in military and naval recruits. These viruses also cause the newly recognized clinical entity pharyngoconjunctival fever occurring as sporadic cases and small outbreaks in the general population (Bell, Rowe, Engler, Parrott & Huebner, 1955), and reported in epidemic form in residential boys' schools in this country (Kendall, Riddle, Tuck, Rodan, Andrews & McDonald, 1957). The occurrence in adults of acute non-bacterial pharyngitis, without conjunctivitis, associated with infection by certain types of adenoviruses was reported by Rowe, Huebner, Hartley, Ward & Parrott (1955) and mainly in children by Ginsberg, Gold, Jordan, Katz, Badger & Dingle (1955) and by Kjellén (1955). Later Roden, Pereira & Chaproniere (1956) showed that infection with Type I adenovirus by nasal instillation gave rise to acute pharyngitis in two out of eleven adult volunteers. Huebner, Rowe, Ward, Parrott & Bell (1954) suggested that the common cold was not caused by members of the adenovirus group, but that Type 3 adenovirus was present as a 'fellow traveller' in some of their cases of common cold, producing infection but not the symptoms in such cases.

A recent trial by Stallones, Hilleman, Gauld & Warfield (1957) with an adenovirus vaccine has shown appreciable protection against severe adenovirus infection in military recruits, the role of the vaccine in protection against mild adenovirus disease in this study being less definite.

The present investigation was made to discover the following points: (i) what proportion of acute respiratory infections occurring in a group of volunteers working in various parts of a teaching hospital were associated with infection by a member of the adenovirus group; (ii) if such infections could be distinguished clinically from those due to other agents; (iii) whether adenovirus vaccines were likely to reduce appreciably respiratory infections in this type of community; (iv) to determine the neutralizing antibody patterns to adenoviruses Types 1–7, 9, and 10 in paired sera from all volunteers, irrespective of whether the current infection was due to an adenovirus.

MATERIALS AND METHODS

Patients studied

Fifty acute respiratory infections were studied in forty-five volunteers, five of the volunteers having two infections during the course of investigation, which ran

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Table 1. *Distribution of acute respiratory infections in forty-five volunteers*

Month	Medical students	Technical staff	Pathologists	Medical officers	Office staff	Ward orderly	Nurses	Total
Oct. 1956	○ ○ ○ ○ ○ (a b) ○ ○ ○ ○	—	—	—	○	—	—	10
Nov. 1956	△ ⊥△ 15	○	○	—	○ △ (c)	—	—	6
Jan. 1957	○ ○ ○	[27]	○ ○ (d) ⊥[19] ⊥△ 24	○	—	—	—	9
Feb. 1957	(b) ○	△ ○ ○ (e)	△	○	—	—	—	6
Mar. 1957	(a) ○	[39] ○	○ ○ (d) [37]	—	(c) ○	○	⊥[41] ○	10
Apr. 1957	○ [45]	○ ○ (e)	—	—	—	—	—	4
May 1957	—	○	—	—	—	—	—	1
June 1957	—	○	—	—	—	—	—	1
July 1957	[GF]	—	—	[49]	—	—	○	3
Totals	19	11	9	3	4	1	3	50

○, Common cold; △, pharyngitis; ⊥△, pharyngitis, adenovirus Type 3 isolated, +serological evidence of adenovirus infection; □, other syndromes; ⊥□, other syndromes+serological evidence of adenovirus infection; [GF], glandular fever; *a*, *b*, *c*, *d*, *e*, volunteers who had two infections; 15, 19, 24, 27, 37, 39, 41, 49=case histories.

from October 1956 to July 1957. Nineteen infections occurred among medical students and three among medical officers working in various departments of the hospital. In addition, there were eleven infections in technicians, nine among pathologists, four among the secretarial staff, three in nurses and one in a ward orderly. The distribution of infections in the different groups of volunteers throughout the period studied, as shown in Table 1, does not represent a single epidemic.

Forty-one of the volunteers were aged between 20 and 36; the remaining four were 19, 45, 54 and 65 years old. Second infections occurred in volunteers aged 21–27.

Clinical methods

Volunteers were requested to report to the laboratory as soon as possible after the onset of upper respiratory symptoms, however mild. At this initial visit the throats of all volunteers were examined, the presence of cervical glands and the appearance of the conjunctivae were noted. Volunteers were examined once only but were asked to fill in a questionnaire which specifically mentioned the order of appearance, type and duration of the following signs and symptoms: sore throat, nasal obstruction, nasal discharge, conjunctivitis, headache, malaise, cough, sputum. Any other signs and symptoms, and morning and evening temperatures were also to be recorded during the course of the acute infection.

Assessment of cases

According to the signs and symptoms occurring in each case, the acute respiratory infections were assigned, where possible, to recognized clinical syndromes.

The common cold was defined as by Stuart-Harris (1955) as a 'short acute disease usually unaccompanied by fever or at the most accompanied by trivial pyrexia and with dominant symptoms of nasal irritation. Sneezing, a clear watery nasal discharge, perhaps slight sore throat and cough usher in acutely a disease which lasts anything from 1–21 days. General symptoms such as malaise usually last only the first day or so, in the later stages the nose is blocked and the voice is nasal. Many individuals regularly experience a more widespread involvement of the respiratory tract than the purely nasal disease of the majority'.

Acute pharyngitis was defined as an acute upper respiratory infection, with or without constitutional symptoms, in which early localizing signs and symptoms were limited to the pharynx and the lymphatic glands draining it. As in the common cold, involvement of the larynx and trachea may regularly occur in some people whatever the aetiology of the pharyngitis.

Cases in this study which could not be classified into the syndromes described above could not be assigned to any other recognized groups, such as acute respiratory disease, bacterial pneumonia, and primary atypical pneumonia.

Laboratory methods

Collection of material and storage. At the initial visit, nose and throat swabs for isolation of haemolytic streptococci, pharyngeal garglings for adenovirus isolation, and 15–20 ml. of venous blood for antibody studies were collected. In the majority

of cases a second specimen of blood was collected 28–31 days later. In forty cases the second specimen was collected after 28–34 days and in seven cases after 36–40 days. In three cases no second specimen was available. The sera obtained from each specimen of blood were stored at −40° C. until sera were available from the second bleeding and each pair could be tested simultaneously. Material for bacterial culture, virus isolation and the 'acute' serum for antibody studies were collected within 1–3 days of the onset of symptoms in thirty-six infections, on the 4th day in six, and on the 5th–8th days after onset in the remaining eight infections. For pharyngeal garglings 10 ml. of fresh HeLa cell culture medium with antibiotics (*vide infra*) were used. The garglings were frozen within 5–30 min. of collection and stored at −40° C.

Bacterial cultures. Nose and throat swabs for the isolation of Lancefield group A, C or G streptococci were cultured aerobically and anaerobically on 5% horse blood agar plates containing 1:500,000 crystal violet.

Tissue cultures. HeLa cell cultures only were employed for adenovirus isolation; the original tissue cultures were received from Dr F. K. Sanders (M.R.C. Virus Research Unit, London School of Hygiene), who also suggested the composition of the nutrient medium in which the cells were grown. Stock cultures were grown directly on glass in stationary 8 oz. Pyrex babies' feeding-bottles with 10 ml. of nutrient medium, and 32 oz. prescription bottles with 25–30 ml. of medium. All cultures were grown at 37° C. with nutrient medium containing 80% Earle's balanced salt solution (Earle, 1943), 10% rabbit serum (inactivated at 56° C. for 30 min.), 5% of an autoclaved 5% solution of lactalbumen hydrolysate (Nutritional Biochemical Corp., Cleveland, Ohio), 5% Hartley's digest broth pH 7·6 (Southern Group Laboratory, Park Hospital, London, S.E. 13), penicillin G 100 units/ml., Streptomycin sulphate 0·2 mg/ml., Neomycin sulphate 0·2 mg/ml., mycostatin 200 units/ml.

Subcultures using the Trypsin technique were made at weekly intervals to further stock cultures and as often as required to replicate cultures (Syverton & Scherer, 1954), in Sanders *PM* 2 bottles with silicone rubber liners.

Batches of HeLa cells were tested at intervals for their sensitivity to the cytopathogenic effect of reference strains of adenovirus Types 1–7, 9 and 10 received from Dr H. G. Pereira, National Institute for Medical Research, Mill Hill. Type 8 was omitted from this study as no respiratory infections without keratoconjunctivitis have been reported with this type, and the batch of HeLa cells in use in this laboratory appeared to be resistant to infection by a known Type 8 adenovirus.

Virus isolation. For virus isolation Sanders *PM* 2 bottles were started with 1 ml. of the above nutrient medium containing about 10^4 cells/ml.; cultures were used for inoculation after 1–3 days incubation at 37° C. horizontally and without rotation. Attempts at isolation of adenovirus were made within 1–14 days of collection of pharyngeal garglings. 0·2 vol. of each gargling were inoculated into at least two Sanders *PM* 2 bottles from which the old medium had been removed. The inoculum was left in contact with the cells at room temperature for 30 min., 1·8 ml. of new nutrient medium was then added and the cultures were incubated at

37° C. Uninoculated cell control cultures were included with each batch of inoculated cultures. Cultures were examined daily, microscopically, for cytopathogenic change. When such change appeared it was allowed to progress until 75% of the cells were involved. 0·5 ml. vol. of fluids from these cultures were inoculated into further new HeLa cell cultures; virus strains isolated in this way were carried through four passages in HeLa cells.

Fluids of cultures inoculated from a single gargling in which no cytopathogenic change was seen after 14–21 days were pooled, spun at 3000 r.p.m. for 20 min. and 0·5 ml. vol. were inoculated into two new HeLa cell cultures which were again observed for not less than 14 days. Cultures showing cytopathogenic change only after the first passage were treated in the same way as the primary cultures. *Cultures in which no change was seen after the first passage were discarded.* Cultures showing doubtful change were passaged until an unequivocal result was obtained. Virus isolation attempts were repeated from all garglings.

Identification of virus strains. Virus strains isolated from pharyngeal garglings were identified as members of the adenovirus group by the characteristic cytopathogenic change accompanied by acid production in HeLa cell cultures (Rowe *et al.* 1955), and the demonstration of adenovirus group-specific antigen by the complement-fixation test (*vide infra*). Strains were typed by serum neutralization tests in HeLa cell cultures using type-specific antisera (Rowe, Hartley & Huebner 1956). Adenovirus strains isolated were kindly typed in parallel by Dr H. G. Pereira.

Serological tests. Paired sera were inactivated at 56° C. for 30 min. and were tested in parallel for complement-fixing antibody to adenovirus group substance and neutralizing antibody to adenovirus Types 1–7, 9 and 10.

Complement-fixation test. Antigen for the complement-fixation test was frequently found to be irreversibly anticomplementary when made from virus grown in HeLa cell cultures with nutrient medium containing rabbit serum. The anticomplementary effect of horse serum was, however, invariably removed after incubation at 56° C. for 30 min.; in cultures used for the preparation of complement-fixing antigen the rabbit serum was therefore replaced by 10% horse serum. For the test, antigen was made from equal parts of cultures of adenovirus Types 3 and 4, the optimal dilution being determined by chessboard titration with a high titre antiserum.

The method used for the complement-fixation test was that of Roden *et al.* (1956) using 'Perspex' haemagglutination plates and a unit volume of 0·025 ml. delivered from a dropper. Twofold dilutions of sera were made in saline.

Neutralization tests. The technique used for the neutralization test was 'Procedure 2' described by Rowe *et al.* (1955). Stock virus preparations from adenovirus Types 1–7, 9 and 10 were prepared and titrated as described by these workers. For the virus titrations and neutralization tests Sanders *PM* 2 bottles were started with 1 ml. of nutrient medium containing approximately 10^5 cells/ml., cultures were used for the tests within 18–24 hr. after preparation. Acute and convalescent sera from all volunteers were screened at a dilution of 1 in 4 Earle's balanced salt solution, for neutralizing antibodies to adenovirus Types 1–7, 9 and 10; antibody titres were subsequently determined using fourfold dilutions of serum.

RESULTS

Clinical findings

The distribution of the different clinical types of acute respiratory infections occurring among the volunteers is shown in Table 1. According to the definition of Stuart-Harris (*vide supra*) thirty-six of the fifty acute respiratory infections studied were the common cold, and all but one of the volunteers in this group remained at work throughout their infection. In twenty-three of these cases the duration of symptoms was 10 days or less, in the other thirteen cases the duration was up to 3 weeks; the symptoms and signs prolonging the course of the illnesses were purulent nasal and post-nasal discharge, the latter frequently accompanied by an irritating cough. In two cases a herpetic eruption on the lips accompanied the onset of nasal symptoms.

One case of infectious mononucleosis, confirmed by the blood picture and positive Paul-Bunnell reaction was found and this reduced the undiagnosed acute respiratory infections to thirteen. Six of these were clinically pharyngitis, all the volunteers in this group remained at work throughout the course of their infections. In four cases the illness was afebrile and very mild, the only symptoms being sore throat and headache lasting from 1–3 days. In the remaining two cases (15 and 24), the pharyngitis was more severe and prolonged and was accompanied by minimal nasal symptoms, clinical details of these are given as strains of adenovirus were isolated from both patients.

Case histories

Case 15. A female medical student, aged 22, presented with sore throat, obstructed left nostril and serous nasal discharge. The nasal symptoms were present on the day of onset only, but the sore throat persisted for 7 days, fluctuating in severity. Her temperature was raised to 99·4° F. on the evening of the second day, returning to normal on the following day. The tonsillar glands, particularly on the left, became enlarged and tender and during the second night of symptoms she was kept awake by pain in the left angle of the jaw, the angle of the left eye, and ache in the left ear. Examination of the throat on the day after the onset of symptoms showed generalized infection of the fauces but no exudate. All symptoms subsided within 8 days.

Case 24. A female pathologist, aged 30, presented with a mild sore throat which became increasingly more painful and persisted for 11 days. A dry non-productive cough was present from the onset of the sore throat and lasted for 16 days. Nasal obstruction without discharge was present from the fourth to the eleventh day of symptoms, there was marked tracheitis from the fourth day accompanied by a husky voice, the latter symptoms persisted for 12 days. The temperature was recorded from the 4th day of symptoms only and was raised to 99° F. on the evenings of the 4th and 5th days, returning to normal the following day. Examination of the throat showed reddening of the fauces but no exudate. All symptoms had subsided after 16 days.

Seven cases remained which did not fall into any particular group. Brief notes

are given below of two of these (Cases 19 and 41), because the results of serological examination suggested the possibility of an adenovirus relationship. The remaining five (Cases 27, 37, 39, 45 and 49), however, showed no evidence of such an association. They did not meet the definitions laid down for the common cold or acute pharyngitis, and they can only be regarded as cases of acute respiratory infection of unknown aetiology. It should be pointed out that although their clinical picture did not suggest virus influenza their sera were not examined for influenza virus antibodies.

Case 19. A male pathologist aged 34 presented with a simple unilateral conjunctivitis accompanied by a cough productive of mucoid sputum. The eye symptoms lasted 2 days, the cough became unproductive on the 6th day but continued for 15 days; on the 2nd day his temperature rose to 100·8° F., returning to normal the following day.

Case 41. A nurse aged 20 had a mild sore throat for 5 days, it increased in severity on the 6th day and was accompanied by nasal obstruction, purulent nasal discharge and a temperature of 99° F. The throat was markedly reddened but there was no exudate, all symptoms had subsided after 11 days.

The clinical types and the intervals between infections, among volunteers who had two acute respiratory infections during the course of the study, are shown in Table 1.

Laboratory findings

Virus isolation. Adenovirus strains were isolated only from the pharyngeal garglings of Cases 15 and 24 both of which clinically were acute pharyngitis with minimal nasal symptoms. Both strains belonged to adenovirus Type 3. In Case 15 the pharyngeal gargling was obtained on the 2nd day of symptoms and virus was isolated consistently on the 10th day after inoculation of primary HeLa cell cultures. In Case 24 the pharyngeal gargling was obtained on the 5th day of symptoms and virus was isolated only after the first blind passage in HeLa cell cultures.

No haemolytic streptococci belonging to Lancefield Groups A, C, or G were isolated.

Serology. In Cases 15 and 24, from which strains of adenovirus Type 3 were isolated, there was also serological evidence of adenovirus infection. In Case 15 the complement-fixing antibody titre rose from 1:2 to 1:16 and there was a rise in neutralizing antibody to Type 3 virus from 0 to 1:4, the interval between sera being 36 days. In Case 24 the complement-fixing antibody titre rose from 1:2 to 1:64 and the neutralizing antibody titre to Type 3 virus rose from 0 to 1:8; there was also a fourfold heterologous response in neutralizing antibody to Type 6 adenovirus, the titre rising from 1:8 to 1:32, the interval between the sera was 28 days. There was also serological evidence of adenovirus infection in two further cases, nos. 19 and 41, in which adenovirus strains were not isolated. In Case 19 a pharyngeal gargling and a conjunctival swab obtained on the 2nd day of symptoms yielded no virus, but there was a rise in complement-fixing antibody titre from $< 1:8$ to 1:16, and a rise in neutralizing antibody to Type 4 virus from 1:8 to 1:64, the interval between sera being 31 days. In Case 41 the pharyngeal gargling was obtained on the 6th day of symptoms and no virus was isolated, the complement-

Table 2. *Studies of neutralizing and complement-fixing antibody to adenovirus Types* 1–7, 9 *and* 10, *in acute and convalescent sera from forty-four volunteers with acute respiratory infections*

(1) All titres are expressed as the reciprocal of the highest original serum dilution giving complete neutralization, and the highest original serum dilution giving 50 % complement fixation.

(2) No neutralizing antibodies to adenovirus Types 9 and 10 were found in any of the sera and they were not examined for neutralizing antibodies to adenovirus Type 8.

(3) *a*, *b*, *c*, *d*, *e*: volunteers who had two infections.

No. of volunteer	Neutralizing antibody to adenovirus types							Complement fixing antibody
	1	2	3	4	5	6	7	
1	0/0	0/0	0/0	0/0	2/2	0/0	2/2	2/2
2	0/0	0/0	0/0	4/4	0/0	0/0	0/0	< 2/< 2
3	32/32	32/< 32	< 8/4	4/4	2/2	32/32	< 8/< 8	4/2
4	0/0	0/0	32/32	64/< 64	0/0	0/0	< 8/< 8	4/4
6*	0/0	0/0	0/0	0/0	2/2	0/0	< 8/< 8	0/0
7*a*	0/0	0/0	0/0	8/< 8	0/0	0/0	< 8/< 8	2/2
8*b*	8/8	2/2	0/0	2/2	2/2	2/2	2/2	16/16
9	0/0	0/0	0/0	0/0	0/0	0/0	2/2	8/< 8
10	8/8	0/0	0/0	0/0	0/0	0/0	2/2	2/2
11	0/0	16/16	0/0	2/2	0/0	2/2	0/0	2/2
12	16/< 8	< 8/< 8	0/0	2/2	0/0	0/0	0/0	2/2
13	0/0	32/32	0/0	0/0	0/0	0/0	0/0	2/2
14	8/8	0/0	8/8	2/2	0/0	0/0	0/0	< 4/< 4
15	32/32	0/2	0/4	0/0	0/0	4/4	32/16	2/16
16*c*	0/0	< 8/< 8	0/0	4/4	0/0	0/0	16/8	8/8
17*d*	0/0	0/0	0/0	8/> 8	0/0	0/0	0/0	0/0
18	2/< 2	32/64	8/16	< 16/< 16	32/32	0/0	< 8/< 8	16/16
19	0/0	0/0	2/2	8/64	2/2	0/0	0/0	< 8/16
20	4/4	0/0	< 8/8	16/16	0/0	< 2/< 2	0/0	4/4
21	< 8/< 8	0/0	0/0	4/4	2/2	0/0	2/2	8/8
22	2/2	0/0	16/8	8/8	128/128	8/8	8/8	16/16
23	2/< 2	8/8	0/0	8/4	0/0	0/0	0/0	8/8
24	< 8/< 8	0/2	0/8	0/2	0/0	8/32	0/0	2/64
25	0/0	8/8	16/16	32/32	0/0	0/0	0/0	64/64
26	0/0	0/0	0/0	0/0	0/0	0/0	0/0	0/0
27	0/0	0/0	0/0	< 8/< 8	32/32	0/0	0/0	0/0
28*e*	32/32	16/16	2/2	2/2	0/0	2/2	0/0	8/8
29	16/16	8/8	< 8/8	2/2	0/0	0/0	0/0	8/4
30*b*†	< 8/< 8	2/2	0/0	2/2	2/2	2/2	0/0	16/16
32	0/0	16/16	8/8	8/8	0/0	0/0	0/0	16/8
33*c*	0/0	> 4/> 4	0/0	4/4	0/0	0/0	> 8/> 8	> 8/> 8
34*a*	0/0	0/0	0/0	4/4	0/0	0/0	2/2	< 2/< 2
35	0/	< 8/	0/	0/	4/	0/	0/	< 2/
36	0/0	0/0	16/16	8/8	0/0	0/0	0/0	2/2
37*d*	0/0	0/0	0/0	8/8	0/0	0/0	0/0	0/0
38	0/0	64/64	0/0	0/0	128/128	0/0	8/8	4/4
39	0/0	0/0	32/32	16/8	0/0	8/8	0/0	8/8
40	32/32	0/0	8/8	2/2	0/0	2/2	2/2	2/2
41	< 2/< 2	0/0	0/4	2/2	0/0	8/64	0/0	< 2/8
42	128/	64/	2/	2/	0/	< 8/	0/	8/
43	0/0	8/8	8/8	< 8/< 8	0/0	0/0	0/0	16/16
44*e*	32/32	16/< 16	2/2	4/4	0/0	2/2	0/0	16/8
45	8/8	0/0	0/0	0/0	0/0	2/2	2/2	0/0
46	< 2/< 2	< 8/< 8	2/2	< 8/< 8	0/0	0/0	0/0	0/0
47	8/8	32/32	0/0	8/16	0/0	0/0	2/2	8/8
48	0/0	0/0	8/8	16/16	0/0	0/0	0/0	2/2
49	0/0	0/0	0/0	8/8	0/0	8/8	0/0	2/2
50	16/16	0/0	0/0	2/2	0/0	< 2/< 2	8/8	0/0
51	0/0	0/0	0/0	< 8/< 8	8/8	0/0	0/0	8/8

* No specimens were available from volunteer no. 5.

† The number 31 was omitted from this series.

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fixing antibody titre rose from < 1:2 to 1:8 and there was a neutralizing antibody rise for Type 3 from 0 to 1:4 and for Type 6 from 1:8 to 1:64. In the remaining forty-six cases there was neither serological nor virological evidence that the illnesses studied were due to adenovirus infection.

The complement-fixing antibody titres to adenovirus group-specific antigen and the neutralizing antibody titres to adenovirus Types 1–7, 9 and 10 in paired sera from forty-seven of the fifty cases of acute respiratory infections are shown in Table 2.

DISCUSSION

There was evidence of adenovirus infection in four of the fifty acute upper respiratory infections studied. In three of these the clinical picture was quite distinct from that of the common cold which accounted for thirty-six of the fifty acute respiratory infections. The two cases of acute non-bacterial pharyngitis (Nos. 15 and 24) which were associated with adenovirus Type 3 infection were clinically very similar to those of Ginsberg *et al.* (1955), although their cases occurred mainly in children and fever was a more prominent symptom. Strains of adenovirus Type 3 were also isolated by these authors and the majority of their cases had a neutralizing antibody rise to adenovirus Type 3 in the convalescent serum. In outbreaks of pharyngoconjunctival fever due to adenovirus Type 3, cases of pharyngitis with fever and cervical lymphadenitis, not accompanied by conjunctivitis, occurred not uncommonly side by side with cases of the fully developed pharyngoconjunctival fever (Bell *et al.* 1955; Kendall *et al.* 1957). In view of the negative bacteriological findings in Cases 15 and 24 and their similarity to cases described by other workers the signs and symptoms in both were considered to be due to infection by adenovirus Type 3.

In Case 19 the main symptoms of simple conjunctivitis, fever, irritating cough and laryngitis form a less well-defined picture. As no adenovirus strain was isolated from this case, definite conclusions cannot be drawn about the infecting adenovirus type from the neutralizing antibody response alone, because Grayston, Loosli, Johnston, Smith & Woolridge (1956) showed that during adenovirus infections occasionally a higher neutralizing antibody response occurred to a heterologous adenovirus type than to the infecting type. Since there was an eightfold rise in neutralizing antibody titre to Type 4 adenovirus in the convalescent serum, this may have been the infecting type. The infection, however, may well have been due to an adenovirus type not included in the antibody studies in this investigation, with a heterologous antibody response to Type 4. The presence of neutralizing antibody to Type 4 adenovirus at a level of 1:8 in the acute serum supports the theory that Type 4 was not the infecting type, since the level of antibody in a serum taken on the second day of symptoms might have been expected to protect against infection with the homologous type of adenovirus (Roden *et al.* 1956).

The fourth case (No. 41), in which there was evidence of adenovirus infection, differed from the clinical picture of the common cold only in the duration of pharyngitis before the onset of nasal symptoms. In this case also the infecting type of adenovirus cannot be stated since no virus strain was isolated. The fourfold rise

in neutralizing antibody titre to adenovirus Type 3 from 0 to 1:4 and the eightfold rise in neutralizing antibody to Type 6 from 1:8 to 1:64, was similar to the antibody pattern in Case 24, in which the infecting strain of adenovirus was Type 3. No definite conclusions can be drawn from these antibody patterns, however, as heterologous responses appear to be variable and unrelated to pre-existing antibody levels in the acute sera of patients with acute respiratory infections due to adenoviruses (Grayston *et al.* 1956). Until there are further studies on sporadic adenovirus infections in adults, it cannot be stated that the symptoms and signs in Cases 19 and 41 were due to adenovirus infection. Adenovirus infection may have been incidental as in some of the cases of common cold transmitted to volunteers by nasal washings (Huebner *et al.* 1954), in which adenovirus Type 3 appeared to be a 'fellow traveller'.

If adenovirus vaccines are effective in preventing the relatively mild adenovirus infections which occurred in this community, then only two or possibly four out of fifty acute respiratory infections could have been prevented.

Studies on the sera of forty-four of the forty-five volunteers, as shown in Table 2, revealed complement-fixing antibody titres to adenovirus group-specific antigen at a level of 1:8 or higher in 16 of these individuals. The highest titre observed in any of the acute sera was 1:64 (Case 25); this was also the titre of the convalescent serum from Case 24. In Cases 15, 24 and 25 sera obtained 8 months after the convalescent bleedings showed that the complement-fixing antibodies had dropped respectively from 1:16 to 1:2, 1:64 to 0 and 1:64 to 1:2. This suggested the possibility that the sixteen volunteers with 'acute serum' antibody levels of 1:8 or higher had had recent adenovirus infections.

In the 'acute serum' specimens of forty-four of the volunteers in my series neutralizing antibody at a level of 1:2 or higher to adenovirus Type 4 was found in 75%. Huebner *et al.* (1954), in a survey of a comparable series of patients without any detectable respiratory infection, found antibodies to Type 4 in about 70%—a figure of the same order.

With regard to adenovirus Type 3 there were 22% with antibodies to this type in Huebner's series as compared with approximately 40% of the 'acute sera' of my own series. If the absence of demonstrable type-specific neutralizing antibody at a level of 1:2 can be accepted as evidence of susceptibility to infection by that type, it can be said that about 60% of the volunteers were at risk to infection with adenovirus Type 3. This antigenic type can generate epidemics, but presumably the contact between the Cases 15 and 24 and the others was not sufficiently close for this.

Although the number of sera examined was small the results probably reflect the frequency of neutralizing antibodies to the different adenovirus types among the staff and student population of a teaching hospital where the majority fall into a comparable age group.

SUMMARY

Four out of fifty acute respiratory infections occurring in forty-five adult volunteers in a teaching hospital, from October 1956 to July 1957, were associated with infection by a member of the adenovirus group. In three of these adenovirus

infections the clinical picture was quite distinct from that of the common cold, but not from other syndromes of unknown aetiology; in the fourth case the clinical picture was not easily distinguishable from the common cold.

It is suggested on the basis of this study that adenovirus vaccines would do little towards reducing sporadic acute respiratory infections in this type of community.

Complement-fixing and neutralizing antibody patterns to adenovirus Types 1–7, 9 and 10 were determined in paired sera from forty-four of the forty-five volunteers.

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