

THE OCCURRENCE OF ADENOVIRUS PRECIPITATING ANTIBODIES IN HUMAN SERA IN IRAN

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Introduction

Since the initial isolation of adenoviruses from children and military recruits with acute respiratory disease (Rowe, *et al*, 1953; Hilleman and Werner, 1954; Huchner *et al*, 1954; Parrot *et al*, 1954) thirty-one serotypes of human adenoviruses have been reported (Gillespie, 1967). These viruses have been associated with various syndromes such as: pharyngo-conjunctival fever, acute febrile conjunctivitis, pneumonia in children and adults, gastero-entritis, mesentric adenitis, myocarditis, nephritis and encephalitis (reviewed by Ginsberg and Dingle, 1965). The only reported isolation of human adenovirus in Iran is that of Naficy *et al* (1967) who recovered adenovirus type 7 from children affected with kerato-conjunctivitis, pneumonitis and diarrhea.

Since a serological survey can provide information concerning the existence of infection in a population, the present serological study has been carried out to determine the incidence of antibodies to the group-reactive precipitating antigen of adenoviruses in human sera in Iran.

MATERIALS AND METHODS

Adenovirus antigen and antiserum. The WBR 1 strain of Bovine Adenovirus 3 (Darbyshire *et al*, 1965) virus received as freeze dried material, was serially passaged in cultures of BHK/21 cells on three occasions. The third passage material was tested for identity against known rabbit antiserum, then used as antigen in all further tests.

Serum samples. The human serum samples were obtained from patients referred to the Workers' Insurance and Koch Diagnostic Laboratories, Tehran, Iran. All the sera were stored at -20° C. until examined by immuno-diffusion tests.

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Immuno-diffusion tests. Immuno-diffusion tests were done by the method of Darbyshire (1962). One percent agar medium containing 0.15 molar sodium chloride and 0.25 percent (w/v) phenol, and buffered to pH 7.2 with 0.1 molar Sorenson's phosphate buffer was prepared and poured into plates. Two sets of wells 0.6cm. in diameter and 0.5cm. apart were arranged hexagonally around a central well, 0.7cm. in diameter. The central well was filled with adenovirus antigen and each of the surrounding wells with test serum. Plates were incubated at between 19°—22° C in a moist atmosphere for 72 hours, then examined for a zone of precipitate under reflected light from a mirror. A given serum considered positive when a distinct precipitation line was observed between the central well and that containing the serum.

RESULTS

The results of immuno-diffusion tests for the presence of group specific antibody of adenoviruses in 322 sera of man are presented in Table I. Some of the results of positive human serum samples arranged according to age grouping are shown in Table II. No significant difference was demonstrated on account of sex or age.

Table I: Results of immuno-diffusion tests for adenovirus precipitation antibody in sera of human.

	Total Sera	No. of Positive	Percentage of Positive
Female	158	31	19.6
Male	164	26	15.8
Total	322	57	17.7

Table II: Results of positive human serum samples according to age groups.

Age (Years)	Percent
Less than 15	16.7
16-45	29.9
Over than 45	19.5

DISCUSSION

Adenoviruses possess a group-reactive antigen which is detected by complement fixation (Hilleman *et al.*, 1955) and gel-diffusion precipitation (Darbyshire, 1964) tests. This antigen has been designated antigen A or L and contains a group-reactive component together with deoxyribonucleic acid (Ginsberg and Dingle, 1965). Serological tests to detect the group specific antibody of adenoviruses are necessarily limited to immuno-diffusion or complement-fixation techniques, in which an antigen prepared from any member of the adenovirus group except the avian strains can be used (Andrewes and Pereira, 1967; Gillespie, 1967). In the present study the Bovine Adenovirus type 3 was used as antigen for the detection of antibodies in the sera of human. From the results of immuno-diffusion tests (Table I) it is evident that adenovirus infection is widespread among the human population in Iran.

The variation in the percentage of the positive serum samples with respect to age groups (Table II) is considered of insignificant value. However, the immuno-diffusion test is generally less sensitive than other serological methods used for demonstration of antibodies to adenoviruses. Thus the overall picture may follow the common pattern of the increase in incidence of antibodies with age (Naito *et al.*, 1962) if all the requisite antigens are available for neutralization or haemagglutination inhibition tests.

Adenoviruses have been well documented as causal agent of human diseases (Ginsberg and Dingle, 1965; Andrewes and Pereira, 1967; Darbyshire and Roberts, 1968) and the present study suggests that these agents may be responsible for both frank and inapparent infections amongst human population of this country.

SUMMARY

By immuno-diffusion tests, 322 serum samples from human were examined for the presence of antibodies to the group-reactive precipitating antigen of adenoviruses. Of the total sera from female, 19.6% and from male, 15.8% had antibody to the group-reactive antigen prepared from Bovine Adenovirus type 3. No significant difference was demonstrated on account of sex or age. The results suggest that adenovirus infection is widespread among the human population in Iran.

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REFERENCES

- Andrewes, C. H. and H. C. Pereira.: Adenoviruses in "Viruses of Vertebrates" Baillièrè, Tindal and Cassell, London, pp. 242-261, (1967).
- Darbyshire, J. H.: Agar Gel Diffusion Studies with a Mucosal Disease of cattle. Res. Vet. Sci. 3, 118-120 (1962).
- Derbyshire, J.H.: The Gel Diffusion Technique in Studies of Viruses of Veterinary Importance. Vet. Bull. Weybridge, 34, 699-705 (1964).
- Darbyshire, J. H. and D.H. Roberts: Some Respiratory Virus and *Mycoplasma* infections of animals. J. Cli. Path. 21 suppl. No. 2, 61-87 (1968).
- Darbyshire, J. H., P.S. Dawson, P.H. Lamont, D.C. Ostier and H.G. Pereira: A new Adenovirus Serotype of Bovine Origin. J. Comp. Path. 75, 327-330 (1965).
- Gillespie, J. H.: Adenoviruses. J. A. V. M. A. 28, 573-575 (1967).
- Ginsberg, H.S. and J.H. Dingle: The Adenovirus group, in "Viral and Rickettsial Infection of Man" edited by Horsfall, F.L. and Tama, I. 4th Edition, J.B. Lippincott, Philadelphia, pp. 860-891 (1965).
- Hilleman, M.R. and J.R. Werner: Recovery of new agent from patients with acute respiratory illness. Proc. Soc. Exp. Biol. Med. 85, 183-188 (1954).
- Hilleman, M.R.: Werner, J.H. and M.T. Stewart: Grouping and Occurrence of RI (Prototype RI-67) Viruses, Proc. Soc. Exp. Biol. Med. 90, 556-562, (1955).
- Huebner, R.J., W.P. Rowe, T.G. Ward, R.H. Parrott and J.A. Bell: Adenoidal-pharyngeal-conjunctival agents. A newly recognized group of common viruses of the respiratory systems, New Engl. J. Med. 251, 1077-1086, (1954).
- Naficy, K., S. Saidi, K. Paya and B. Khosravi: Outbreak of Adenovirus Infection in Children's Hospital, presented in the Meeting of Iranian Pediatric Society, Ramsar, Iran (1967), in Persian.
- Naito, H., T. Goto, H. Nakamura, H. Shidda and M. Matumoto: Adenovirus Infections as Studied by Complement Fixation, Jap. J. Exp. Med. 32, 323-334, (1962).
- Parrott, R.H., W.P. Rowe, R.J. Huebner, H.W. Bernton and N.B. McCullough: Outbreak of febrile pharyngitis and conjunctivitis associated with type 3 adenoidal-pharyngeal conjunctival virus infection. New Engl. J. Med. 251 1087-1090, (1954).
- Rowe, W.P., R.J. Huebner, L.K., Gilmor, R.H. Parrott and T.G. Ward: Isolation of a cytopathogenic agent from human adenoids undergoing spontaneous degeneration in tissue culture, Proc. Soc. Exp. Biol. Med. 84, 570-573, (1953).