Antineuraminidase Antibody Response to Vaccination of Chickens with Intact Virus and Different Subunit Preparations of the Influenza Virus Strains A/Sing/1/57 (H2N2), A/Hong Kong/1/68 (H3N2) and A/Port Chalmers/1/73 (H3N2)*

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Summary

The antineuraminidase (AN) antibody response to vaccination of chickens with intact virus and different subunit preparations of the influenza virus strains A/Sing/1/57 (H2N2), A/Hong Kong/1/68 (H3N2) and A/Pt. Chalmers/1/73 (H3N2) was tested comparatively.

Using a photometric method capable of analysing mixtures of AN antibodies against antigenically different N2 neuraminidases, it was concluded that vaccination with subunits produced by treatment with bromelain and Sarkosyl can yield AN antibody response against heterologous neuraminidase. By contrast, vaccination with intact and ether-treated virus gave AN antibody response against homologous neuraminidase, only.

These findings were confirmed when testing sera by means of enzyme inhibition test and by adsorption experiments with homologous and heterologous neuraminidases.

The conclusion was reached that the NA's of the strains A/Sing/1/57 and A/Pt. Chalmers/1/73 share antigenic determinants and that the NA of the strain A/Hong Kong/1/68 shares antigenic determinants with that of the strains A/Sing/1/57 and A/Pt. Chalmers/1/73.

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Abbreviations used: ACU = antibody concentration unit; AN = antineuraminidase; HA = hemagglutinin; HCU = hemagglutinin concentration unit; mU = micro units; NA = neuraminidase; NIT = neuraminidase inhibition test; PBS = 0.15 M NaCl solution buffered at pH 7.0 with 0.01 M phosphate.
Introduction

The hemagglutinins (= HA's) and neuraminidases (= NA's) of influenza viruses show independent antigenic variation (20, 23) which has been assumed to result from recombination between strains of human and animal origin (15, 23).

Another possible explanation is to assume that antigenic variation occurs by rearrangement of antigenic components common to a family of strains and that some of these antigens occupy an inaccessible subsurface position (4).

If this were so, disruption of virus could unmask such antigens.

This has been repeatedly described for the HA's of different influenza virus A strains produced by ether-treatment (4, 10).

Furthermore, it has been previously reported (5) that influenza virus N2 NA's isolated by use of bromelain-treatment can differ antigenically from the NA present on intact virus.

Therefore, the experiments described in this paper were designed in order to examine whether or not vaccination of chickens with viral subunits produced by different techniques can yield AN antibody response against heterologous NA.

The NA's of influenza virus H2N2 and H3N2 strains have been reported to be antigenically inhomogeneous (6, 12, 17).

The antigenic relationship between N2 NA's has been examined by means of a photometric hemagglutination inhibition test (ACU-test) (8), using recombinants possessing a relevant NA and an irrelevant HA (12).

It was found that three classes of N2 NA, designated NAa, NAb and NAe can be defined: NAa is represented by the NA of the strain A/Sing/1/57 and other H2N2 strains, NAb by the NA of A/Hong Kong/1/68 virus and NAe by the NA of A/Port Chalmers/1/73 virus (12). This classification agrees with previous reports (17).

Each class of AN antibodies gave different titer ratios when allowed to react with the recombinants A/Bel/34 (H0) — A/Sing/57 (N2) (= Bel-Sing), A/equine—Prague (Heq1) — A/Hong Kong/1/68 (N2) (= X15—HK) and mixtures of both recombinants (12).

It has been found that recording these titer ratios for reference sera and test sera can be utilized to determine the concentration of the different classes of AN antibodies in the sera to be tested (12).

This technique has been found to give reliable results when used for analysing artificially prepared mixtures of AN antibodies (12).

This method was used for recording comparatively the AN antibody response to vaccination with intact virus and different subunit preparations of the influenza virus strains A/Sing/1/57 (H2N2), A/Hong Kong/1/68 (H3N2) and A/Port Chalmers/1/73 (H3N2). In addition, sera were tested by means of NIT (1) and by examining the influence of adsorption with homologous and heterologous NA on their AN titers.

Materials and Methods

Virus

The strains of egg-adapted non-inactivated influenza virus employed were:
A/Sing/1/57 (H2N2) (= A/Sing), A/Hong Kong/1/68 (H3N2) (= A/HK), A/Pt. Chalmers/1/73 (H3N2) (= A/PC) and the recombinants A/Bel/34 (H0)—A/Sing/1/57