AN OUTBREAK OF INFLUENZA A (H3N2) IN ACCRA IN 1996

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SUMMARY
During the rainy season of 1996 there were reports of an unusual febrile illness thought to be malaria and/or “flu” in Accra, Ghana. Thick and thin smears from finger prick samples and 10ml venous blood were taken from sixty-two (62) patients for malaria microscopy and screening for antibodies against four strains of influenza A and one strain of influenza B respectively. Fifty-four percent (54%) of the patients were positive for malaria parasites whilst all the patients had high hemagglutination inhibiting antibody titres for Influenza A/Wuhan/359/95 (H3N2). The results indicate that the illness was due to an outbreak of influenza A/H3N2 strain with some of the patients having active malaria.

Keywords: Influenza A, Malaria, Upper Respiratory Tract Infection, Rainy Season

INTRODUCTION
Upper respiratory tract infections (URTI) are the second most common clinical diagnosis after malaria which is endemic in Ghana. Upper respiratory tract infections occur throughout the year but peak during the dry, harmattan season from November to February. Malaria transmission also occurs throughout the year with peaks during and immediately after the rainy season, June to September.

During the rainy season of 1996, June to September, there were anecdotal reports from health providers in Accra of an unusual febrile illness thought to be malaria and/or “flu”. There was public concern about the illness as well, as reported in the lay press and some speculated that the illness might be due to chloroquine resistant malaria parasites because such resistance has been reported in Ghana since 19871,2.

Most respiratory tract infections are believed to be viral in origin. There are however only a few sero-epidemiological studies done to identify viruses responsible for respiratory tract infection in Ghana. Pasca and Amoah3 confirmed the presence of complement fixing antibodies to respiratory viruses in Ghanaian sera and so did Minami et al4 who published a preliminary study on viral infections using some Japanese strains of the Hong Kong influenza A virus. Nowacki and Addy5 reported increased respiratory tract infections in the month of October 1973 that was later confirmed virologically to be due to the Hong Kong influenza A virus6. This therefore indicated that apart from the influenza pandemic of 1968, there had been an epidemic of influenza A in Accra in 1973 which was documented virologically. This report is on a serological investigation undertaken to determine the role of influenza viruses in the febrile illness which occurred during the rainy season of 1996.

PATIENTS AND METHODS
Patients seen over a one month period (21st August - September 1996) by one of us at the Department of Child Health and adult patients seen at the Korle Gonno Urban Health Centre with clinical diagnosis of malaria, respiratory tract infection or both were

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included in the study. Children with cerebral malaria were excluded. Thick and thin blood smears from finger prick samples and 10 ml of venous blood were obtained from patients for malaria microscopy and screening for Influenza A and B strains respectively. All the serum samples for viral serology were unpaired.

Viral serology was carried out using the hemagglutination inhibition test with unpaired sera. Four strains of Influenza A and one strain of Influenza B were used. The virus strains were as follows, Influenza A/Texas/36/91 H1N1, Influenza A/Wuhan/371/95 H1N1, Influenza A/Thessalonika/1/95 H3N2, Influenza A/Wuhan/359/95 H3N2 and Influenza B/Harin/7/94. These viruses had been grown in eggs and were donated by the WHO Collaborative Centre for Reference and Research on Influenza, London, England.

All the serum samples were treated to remove non-specific inhibitors found in normal sera. The combination of heat, trypsin and periodate treatment was adopted. Commercial crystalline trypsin (Difco 1:250) was dissolved in 0.1 M PBS pH 8.2 in a concentration of 8mg/ml. One half volume of the trypsin solution was added to one volume of the serum and the mixture was immediately heated at 56 degrees C for 30 minutes in a water-bath. The mixture was then cooled to room temperature (RT). Three volumes of aqueous 0.01 M potassium periodate solution were added and the mixture was held at least 15 minutes at room temperature. Three volumes of 1% glycerol saline solution was added. The serum at this stage had been diluted 7.5. An additional 2.5 volumes of saline solution were added to bring the dilution to 1:10.

To carry out the hemagglutination inhibition test, 2-fold serial dilutions of treated sera were made starting from 1:20 in microtitre plates. Virus suspensions containing 4 haemagglutinin units (4HAU) were added to each dilution. After incubation at RT for 30 minutes, suspension of day-old chick RBC’s was added and incubated further for 30 minutes at RT. The titre was considered as the reciprocal of the initial dilution of serum which completely inhibited agglutination.

RESULTS
Forty-four (44) samples were obtained from the Department of Child Health and 18 samples from the Polyclinic. Clinical and haematological features of the patients from the Department of Child Health will be reported separately. There were 38 males and 24 females aged between nine months and 42 years. Thirty-four (54.8%) of them had malaria parasites in their peripheral blood film. The only species identified was Plasmodium falciparum. None of the patients had a titre greater than 1:40 for the two H1N1 strains of influenza A and only three (3) of the samples had titres of 1:80 with the Influenza B strain. All patients had markedly elevated titres against the two H3N2 strains of influenza A (see Table 1). Fifty out of 62 samples had titres 1:640 to Influenza A/Wuhan/359/95 H3N2 whilst 24 out of 62 had similar titres to Influenza A/Thessalonika/1/95 H3N2. The overall geometric mean titre for the A/Wuhan/359/95 H3N2 strain was 1026.6 and 489.6 for the A/Thessalonika/195 H3N2. The titres for the other strains were generally 1:20 or less.

DISCUSSION
The results of the hemagglutination inhibition tests indicate strongly that there was an outbreak of infection by an Influenza A H3N2 strain during the rainy

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<th>Table 1: Antibody Titres of the Various Strains of Influenza Virus Tested by Haemagglutination Inhibition.</th>
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<td>Influenza Strain</td>
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<tr>
<td>A/Wuhan/359/95 H3N2</td>
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<td>A/Texas/36/91 H1N1</td>
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<td>A/Wuhan/371/95 H1N1</td>
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period of 1996. Typical post-infection titres are 1:40 and immunity to influenza is predominantly strain specific and does not last long. The high geometric mean titre of 1026.6 for Influenza A/Wuhan/359/95 H3N2 strain therefore indicate that the outbreak was due to that strain or a very closely related strain.

Information on prevalent strains is important in determining the composition of influenza vaccines for each anticipated outbreak. This is particularly important in temperate climates where epidemics tend to occur during the winter months which may prove fatal to vulnerable groups. Such information on strain(s) circulating in tropical countries like Ghana is equally important since tourists may also wish to take advantage of use of current vaccines if they are visiting areas with increased respiratory infections due to influenza. The Influenza A/Wuhan strain is part of the current vaccine composition. Influenza B, unlike Influenza A does not usually give rise to pandemic. Previous documented outbreaks of influenza have occurred at about the same time in the year as reported in this paper. Rapid spread of the virus during this time may be attributed to the cold weather associated with the rains and overcrowding in sleeping facilities.

In this serologic investigation, unpaired sera were used instead of paired sera of acute and convalescent phase samples. The sera were all taken at the first visit to the clinic and the high titres registered shows that the patients were in the early convalescent stage at the time that they sought medical help which has been the practice in most febrile viral diseases. Addy et al (1976) were also unable to obtain paired sera from the general population and succeeded only with the medical students and junior doctors who presented with the disease.

In our setting it is important to establish the strains of influenza virus outbreaks by both serological as well as viral isolation techniques since mixed infection with malaria may affect clinical judgement. Fifty five percent (54.8%) of the patients had malaria parasites in their peripheral blood film as well as high titres of H3N2 antibodies. Investigation of outbreaks of the illnesses as experienced during the period under this study may be done by making facilities for these tests available and also setting up surveillance systems using outpatient and school health facilities. This system will then provide much needed epidemiological information of global interest and also provide practical indicators of the causation of unusual febrile illness with a respiratory component.

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REFERENCES


