110. Johnson, W. J., and Corte, G. Inhibition of isoniazid acetylation *in vitro* and *in vivo*. *Proc. Soc. Exper. Biol. & Med.* 92:446-448, 1956.
111. Davis, B. D., and Maas, W. K. Analysis of biochemical mechanism of drug resistance in certain bacterial mutants. *Proc. Nat. Acad. Sc.* 38:775-785, 1952.
112. Biehl, J. P., and Vilter, R. W. Effect of isoniazid in vitamin B_6 metabolism: its possible significance in producing isoniazid neuritis. *Proc. Soc. Exper. Biol. & Med.* 85:389-392, 1954.
113. Yoneda, M., and Asano, N. Competitive action of isonicotinic acid hydrazide and pyridoxal in amino acid decarboxylation of *Escherichia coli*. *Science* 117:277-279, 1953.
114. Meister, A., and Downey, P. F. Evidence for participation of vitamin B_6 in glutamine-α-keto acid transamination-deamination reaction. *Proc. Soc. Exper. Biol. & Med.* 91:49-52, 1956.
115. Davison, A. N. Mechanism of inhibition of decarboxylases by isonicotinyl hydrazide. *Biochim. et biophys. acta* 19:131-140, 1956.
116. Ungar, J., Tomich, E. G., Parkin, K. R., and Muggleton, P. W. Effect of pyridoxine on action of isoniazid. *Lancet* 2:220, 1954.
117. Zatman, L. J., Kaplan, N. O., Colowick, S. P., and Ciotti, M. M. Isolation and properties of isonicotinic acid hydrazide analogue of diphosphopyridine nucleotide. *J. Biol. Chem.* 209:467-484, 1954.
118. *Idem.* Effect of isonicotinic acid hydrazide on diphosphopyridine nucleotidases. *J. Biol. Chem.* 209:453-466, 1954.
119. Schaefer, W. B. Effect of isoniazid on dehydrogenase activity of *Mycobacterium tuberculosis*. *J. Bact.* 79:236-245, 1960.
120. Russe, H. P., and Barclay, W. R. Effect of isoniazid on lipids of tubercle bacillus. *Am. Rev. Tuberc.* 72:713-717, 1955.
121. Boone, I. U., and Woodward, K. T. Relationship of pyridoxine and its derivatives to mechanism of action of isoniazid. *Proc. Soc. Exper. Biol. & Med.* 84:292-296, 1953.

MEDICAL INTELLIGENCE

HUMAN INFLUENZA INFECTION WITH PROVED VIREMIA*

Report of a Case

KIARASH NAFICY, M.D.†

BOSTON

ALTHOUGH there is some indirect evidence in the medical literature that viremia may occur during human influenza infections[1,2] the isolation of this virus from a patient's blood has not, to the best of my knowledge, been reported. The present communication describes the isolation of influenza virus Group A, Type 2, from both the blood and throat secretions of a patient with clinical manifestations of influenza.

CASE REPORT

Three days before admission a 40-year-old physician noted the onset of severe headache and generalized malaise. He did not believe that he was febrile. This continued until the day before admission, when he noted shaking chills, and the temperature rose to 104°F. At that time he felt confused and somewhat restless. On the morning of admission to the Peter Bent Brigham Hospital he had several bouts of shaking chills followed by fever.

At 32 years of age an episode of fever accompanied by chest and arm pain had resulted in hospitalization and a diagnosis of idiopathic pericarditis. At the time of discharge from the hospital the electrocardiogram had returned to normal. No immunization against influenza had been taken at any time.

Physical examination revealed no abnormalities other than an enlarged thyroid gland. Throughout the hospital course the lungs were clear to percussion and auscultation, and the heart sounds were normal, without any murmur or rub.

*From the Research Division of Infectious Diseases, Children's Hospital Medical Center, and the departments of Pediatrics and Medicine, Harvard Medical School and Children's Hospital Medical Center.

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†Public Health Service international postdoctoral fellow, National Institutes of Health, Public Health Service, United States Department of Health, Education, and Welfare.

The white-cell count was 10,650, with 60 per cent neutrophils, 23 per cent band forms, 15 per cent lymphocytes, 1 per cent monocytes and 1 per cent basophils. The hematocrit was 51.5 per cent, and the corrected erythrocyte sedimentation rate 4 mm. per hour. Throat culture grew alpha-hemolytic streptococci and *Diplococcus pneumoniae*. Sputum culture revealed alpha-hemolytic streptococci, *D. pneumoniae*, *Staphylococcus aureus* and *Escherichia coli*. Febrile agglutination tests, including typhoid O and H, paratyphoid A and B, *Brucella abortus* and *Proteus X-19*, were negative.

The temperature on admission was 99.4°F., and on the following day the patient experienced shaking chills and the temperature rose to 103.6°F. On the 3d and 4th days the temperature did not rise over 100.2°F., but on the 5th day he again had a shaking chill accompanied by a rise in temperature to 102.8°F. From the 6th day on, he had only low-grade fever and noted a gradual diminution of the marked malaise. He became completely afebrile on the 9th hospital day. An electrocardiogram obtained on the 3d hospital day showed flattened T waves in Lead V_6. A chest film on the 4th day revealed that the heart was somewhat enlarged in transverse diameter as compared with films taken after the previous bout of pericarditis. Fluoroscopy on the 6th day showed enlargement of the left ventricle. Five days later an electrocardiogram revealed a normal T wave, and a chest film showed normal cardiac size and shape.

On the 2d hospital day specimens were obtained for attempts at viral isolation. These included clotted blood, a throat wash and a stool specimen that was made up as a 10 per cent suspension in tissue-culture medium.

Aliquots of each of these specimens (0.2 ml.) were inoculated into both primary grivet monkey-kidney and human-amnion cell cultures as well as into the amniotic sac of 7-day-old embryonated hens' eggs. Inoculated cultures and uninoculated controls were tested for hemadsorption 8 or 10 days after inoculation. The amniotic fluid of inoculated and control eggs was harvested on the 5th to the 7th day after inoculation and tested for hemagglutinating factors. Table 1 summarizes the results of the laboratory tests involved in the original isolation, passage and reisolation of the agent.

Hemagglutination-inhibition tests with guinea-pig red cells were used as a means of identifying the agent present in amniotic fluid that had been inoculated with passage material. Tests in accordance with the hemagglutination-inhibition technic recommended by the Committee on Standard Serological Procedures in Influenza Studies[3] revealed no inhibition with antibody to prototype influenza A (PR-8) and B (Arizona) strains. Antiserum to influenza Group A, Type 2 (Asian), inhibited hemagglutination by both the blood and the throat agent in dilutions up through 1:180. Subsequently, the hemagglutinating agent thus identified was reisolated in eggs from both blood and throat specimens (Table 1).

Using blood obtained on admission as an acute-phase and blood obtained 2 weeks after admission as a convalescent-phase specimen, an assay was done for hemagglutination-inhibition antibody. The blood agent, throat agent and a standard strain of influenza virus Group A, Type 2, were all used as antigens. The acute-phase blood in dilutions as low as 1:5 did not inhibit hemagglutination with any of the 3 agents, whereas the convalescent-phase serum inhibited hemagglutination by blood, throat and standard antigen against influenza virus Group A, Type 2, in dilutions through 1:80.

TABLE 1. *Summary of Attempts to Isolate Influenza Virus from Various Materials.*

MATERIAL	RESULTS OF 1ST ATTEMPT*						RESULTS OF 2D ATTEMPT				
	ORIGINAL INOCULATION			1ST PASSAGE			ORIGINAL INOCULATION		1ST PASSAGE		
	human amnion	*grivet monkey kidney*	*egg*†	*human amnion*	*grivet monkey kidney*	*egg*†	*grivet monkey kidney*	*egg*†	*grivet monkey kidney*	*egg*†	*identification*‡
Throat	–	–	+	–	+	+	–	+	+	+	Influenza virus Group A, Type 2
Blood	–	+	+	–	+	+	+	+	+	+	Influenza virus Group A, Type 2
Stool	–	–	–	–	–	–	–	–	–	–	
Controls	–	–	–	–	–	–	–	–	–	–	

*Virus recovered.
†7-day-old embryonated hens' eggs, inoculated intra-amniotically.
‡By hemagglutination-inhibition tests.

An attempt was made to determine the amount of virus present in both the blood and throat specimens. Serial half-log dilutions were made of both specimens and then inoculated into eggs. Only the undiluted specimens were positive.

DISCUSSION

Repeated isolation of influenza virus Group A, Type 2, from a specimen of this patient's blood gives clear evidence that on occasion viremia may occur in influenza caused by this agent. A report of the detection of influenza virus from the liver, spleen, kidney, heart and lymph nodes of patients who died during the outbreak of Asian influenza in 1957[1] strongly suggests that the virus might enter the circulation during the course of the disease. The report of isolation from human urine by another investigator[2] affords additional evidence that viremia may occur. Hamre, Appel and Loosli[4] have shown that viremia may be established in mice after intranasal inoculation of influenza virus Group A (PR-8). A low titer of virus was sporadically demonstrable in the blood only of mice that had a high viral concentration in their lungs. These investigators suggested that viremia in mice might arise as a result of a spillover from the pulmonary focus. If one accepts such a mechanism in human influenza infection, it seems logical to look for viremia at the peak of pulmonary infection rather than at an earlier stage.

Loosli and his co-workers[5] have shown that in mice given airborne influenza infection, both pneumonia and antibody to the agent develop. When mice are given influenza antibody intraperitoneally at the time of viral inoculation pneumonia but not active immunity develops. These results are interpreted by Hamre, Appel and Loosli[4] as indicating a need for generalized spread of virus to antibody-forming sites before active immunity can occur. If this assumption is correct and if it also applies to human influenza infections one can hypothesize that viremia of at least some degree occurs in all patients with influenza infection in whom antibody to the agent develops.

To my knowledge there have been no previous reports of the isolation of influenza virus from the blood of patients. Two papers[6,7] have noted unsuccessful attempts at such isolation. The possibility also exists that there are many unpublished accounts of other unsuccessful attempts at such isolation. One unpublished study by Gresser and Dull[8] includes 9 patients with the clinical signs and symptoms of influenza, with isolation of influenza virus from the throat washings of 7 and without isolation of the virus from any of the blood specimens when the washed leukocyte fractions were tested. It is difficult to account for the differences between the present case and the previous cases studied. As previously suggested,[7] the viremia in influenza may be quite transient, and by chance the present specimen was obtained at the proper time. Another unlikely possibility is that the present patient had some immunologic defect. However, both this patient and those studied by others[7] had no detectable hemagglutination-inhibiting antibody at the time blood was drawn for viral studies. Furthermore, medical history, antibody response to the agent isolated and serum electrophoretic pattern give no indication of any abnormalities of the present patient's immune mechanism. Minuse and his associates[7] suggest that nonspecific inhibitors in the patients' blood may have accounted for their failure to demonstrate influenza virus in blood specimens. The possible lack of such inhibitors was not investigated in the present patient.

SUMMARY

Influenza virus Group A, Type 2, was isolated and reisolated from both the throat washings and blood specimens of a forty-year-old physician hospitalized with shaking chills and fever. A significant rise in hemagglutination-inhibiting antibody was demonstrated both to the agent isolated from the patient and to the standard influenza antigen. Although the report of isolation of influenza virus at autopsy from many of the organs of influenza patients gives evidence of a viremia phase in human influenza, the present study is believed to be the first report of a direct isolation of influenza virus from a patient's blood.

REFERENCES

1. Kaji, M., Oseasohn, R., Jordan, W. S., Jr., and Dingle, J. H. Isolation of Asian virus from extrapulmonary tissues in fatal human influenza. *Proc. Soc. Exper. Biol. & Med.* 100:272-275, 1959.

2. Zakstel'skaya, L. Y. Recovery of virus from urine of patients with epidemic influenza. *Gripp i OKVDP. Tr. ob'yed. sess. AMN SSSR. Moscow*, p. 72, 1953.
3. Committee on Standard Serological Procedures in Influenza Studies. Agglutination-inhibition test proposed as standard of reference in influenza diagnostic studies. *J. Immunol.* 65:347-353, 1950.
4. Hamre, D., Appel, J., and Loosli, C. G. Viremia in mice with pulmonary influenza A virus infection. *J. Lab. & Clin. Med.* 47: 182, 1956.
5. Loosli, C. G., Hamre, D., and Berlin, B. S. Air-borne influenza virus A infections in immunized animals. *Tr. A. Am. Physicians* 66:222-230, 1953.
6. Kilbourne, E. D. Studies on influenza in pandemic of 1957-1958. III. Isolation of influenza A (Asian strain) viruses from influenza patients with pulmonary complications: details of virus isolation and characterization of isolates, with quantitative comparison of isolation methods. *J. Clin. Investigation* 38:266-274, 1959.
7. Minuse, E., Willis, P. W., III, Davenport, F. M., and Francis, T., Jr. Attempt to demonstrate viremia in cases of Asian influenza. *J. Lab. & Clin. Med.* 59:1016-1019, 1962.
8. Gresser, I., and Dull, H. B. Personal communication.

BRIEF RECORDING

Hemolytic Reaction after Novobiocin Therapy

CAPTAIN JOHN R. MONTGOMERY, M.C., USAR*

FORT RUCKER, ALABAMA

A SIX-year-old girl was seen at the United States Army Hospital, Fort Rucker, Alabama, with a chief complaint of mild jaundice and dark urine present for one day. The child had previously been seen by a civilian physician, five days before admission, because of sore throat and fever, with a maximum temperature of 105°F. At that time she had been thought to have pharyngitis, and novobiocin, 30 mg. per kilogram of body weight per day, was started by mouth. The fever subsided and the patient improved. However, on the day of admission she was noted to be mildly jaundiced and had been passing dark-brown urine.

Physical examination disclosed icteric sclerae and pale mucous membranes and conjunctivas. The throat was red, but no exudate was present. The remainder of the physical examination was negative.

The initial impression was that of hepatitis. A blood specimen revealed marked hemolysis on three different occasions, and a hemolytic reaction was suspected. The initial white-cell count was 4800, with a normal differential. The hemoglobin was 7.0 gm. per 100 ml., and the reticulocyte count 1.1 per cent. The blood urea nitrogen was 24.2 mg. per 100 ml. The remainder of the blood chemical findings, including the antistreptolysin-O titer, were within normal limits. A red-cell fragility test showed hemolysis at 0.50 per cent and ending at 0.00 per cent. A tourniquet test was negative. The platelet count was normal. Blood cultures were negative at ten days. During the first twenty-four hours in the hospital the hemoglobin dropped to 4 gm. per 100 ml. The urine was within normal limits except for a trace of bile and a positive test for hemoglobin. The direct and indirect Coombs tests were positive. The blood was Type O+, and two transfusions of this type of blood were given. Prednisone (Meticorten), 40 mg. per day, was started. After the two blood transfusions the hemoglobin rose to 10.2 gm. per 100 ml. The reticulocyte counts increased steadily from 1.1 per cent to a high of 10.2 per cent just before discharge on the fifteenth hospital day. The hemoglobin rose slowly from 10.2 gm. per 100 ml. after transfusions to a discharge level of 13 gm.

The urine cleared within two days after transfusion and institution of cortisone therapy, and the patient became essentially asymptomatic. She was discharged on the fifteenth hospital day with a final diagnosis of acquired hemolytic anemia.

It is possible that the hemolysis resulted from sepsis, but this is unlikely in view of the normal white-cell counts and the absence of fever during the hospital stay. This hemolytic reaction could also have been of the idiopathic variety, but novobiocin remains strongly suspected as the etiologic agent.

The patient has been seen on several follow-up visits, and the hemoglobin is holding steady at 13.5 gm. per 100 ml. The Coombs tests, direct and indirect, have returned to negative, and she is doing well.

*Member, Pediatric Clinic, United States Army Hospital.

BY THE LONDON POST

Lord Nuffield — Pharmacy in Britain — Holiday Reading

JOHN LISTER, M.D.

LONDON, ENGLAND

THE story of the life of Lord Nuffield, who died in August at the age of eighty-six, is like a fairy tale. Born in 1877, William Morris was educated in local schools until the age of sixteen, when, having shown some mechanical aptitude, he was sent to work in a bicycle shop in Oxford. Within a year he had borrowed £4, with which he opened a shop on his own account. He started by repairing bicycles, then he sold and raced them, and later he produced a model of his own. In his first six years of bicycle manufacturing he accumulated £2,000 of capital, and in another ten years, by the age of thirty-three, he had doubled that amount. By 1911 there were some 50,000 private motorists in Britain, and in the following year the Morris car appeared. During World War I the Morris works were turned over to war work, but at the end of hostilities motorcar production was started in earnest. In 1922 nearly 7000 cars were sold, and by 1925 the annual figure had risen to over 52,000.

His business success thus assured, he began to direct his attention to giving financial support to advance the study and practice of medicine, which, in fact, had secretly been his own first choice of career.