Epidem. Inf. (1987), **99**, 5–54
Printed in Great Britain

A new concept of the epidemic process of influenza A virus

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(*Accepted 2 March 1987*)

SUMMARY

Influenza A virus was discovered in 1933, and since then four major variants have caused all the epidemics of human influenza A. Each had an era of solo world prevalence until 1977 as follows: H0N1 (old style) strains until 1946, H1N1 (old style) strains until 1957, H2N2 strains until 1968, then H3N2 strains, which were joined in 1977 by a renewed prevalence of H1N1 (old style) strains.

Serological studies show that H2N2 strains probably had had a previous era of world prevalence during the last quarter of the nineteenth century, and had then been replaced by H3N2 strains from about 1900 to 1918. From about 1907 the H3N2 strains had been joined, as now, by H1N1 (old style) strains until both had been replaced in 1918 by a fifth major variant closely related to swine influenza virus A/Hswine1N1 (old style), which had then had an era of solo world prevalence in mankind until about 1929, when it had been replaced by the H0N1 strains that were first isolated in 1933.

Eras of prevalence of a major variant have usually been initiated by a severe pandemic followed at intervals of a year or two by successive epidemics in each of which the nature of the virus is usually a little changed (antigenic drift), but not enough to permit frequent recurrent infections during the same era. Changes of major variant (antigenic shift) are large enough to permit reinfection. At both major and minor changes the strains of the previous variant tend to disappear and to be replaced within a single season, worldwide in the case of a major variant, or in the area of prevalence of a previous minor variant.

Pandemics, epidemics and antigenic variations all occur seasonally, and influenza and its viruses virtually disappear from the population of any locality between epidemics, an interval of many consecutive months. A global view, however, shows influenza continually present in the world population, progressing each year south and then north, thus crossing the equator twice yearly around the equinoxes, the tropical monsoon periods. Influenza arrives in the temperate latitudes in the colder months, about 6 months separating its arrival in the two hemispheres.

None of this behaviour is explained by the current concept that the virus is surviving like measles virus by direct spread from the sick providing endless

chains of human influenza A. A number of other aspects of the human host influenza A virus relationship encountered in household outbreaks are among the list of 20 difficulties that are inexplicable by the current concept of direct spread.

Alternative concepts have usually been designed to counter particular difficulties and are incompatible with other features of influenzal behaviour in mankind. The new concept detailed in the appendix provides simple explanations for most if not all of the difficulties. It proposes that influenza A virus cannot normally be transmitted during the illness because it too rapidly becomes non-infectious in a mode of persistence or latency in the human host. Many months or a year or two later it is reactivated by a seasonally mediated stimulus which, like all seasonal phenomena, is ultimately dependent on variations in solar radiation caused by the tilt of the plane of earth's rotation in relation to that of its circumsolar orbit. The carriers, who are always widely seeded throughout the world population, become briefly infectious and their non-immune companions, if infected, comprise the whole of the next epidemic. The reactivated virus particles must encounter the immunity they have engendered in the carrier, thus allowing minor mutants an advantage over virions identical with the parent virus, and so favouring antigenic drift and automatic disappearance of predecessor and prompt seasonal replacement. Antigenic shift and recycling of major variants may also be explained by virus latency in the human host.

INTRODUCTION

Davenport has reminded us that 'Epidemiological hypotheses must provide satisfactory explanations for *all* the known findings – not just for a convenient subset of them' (Davenport, 1977). The current concept of the epidemiology of influenza A fails lamentably to pass Davenport's test. The common belief among doctors and their patients is that influenza virus, like measles virus, is surviving and causing epidemics in mankind by endless chains of direct transmissions of which each link in the chain is a person sick with influenza. Numerous observations and laboratory findings, some of which will be discussed, make the 'measles model' untenable for human influenza. The discovery that influenza A viruses parasitize many non-human hosts, both wild and domesticated, has added to the complexity of influenza epidemiology and promoted valuable epidemiological speculations, some of which will be considered in relation to the new concept of epidemic process advanced in this paper.

The new concept, detailed in the Appendix, suggests that human influenza A virus cannot usually be transmitted by the patient during his attack of influenza because it too rapidly undergoes changes leading to prolonged innocuous residence in his tissues. Next season or later, in response to a seasonally mediated stimulus, the virus is reactivated so that the carrier, though not again ill, becomes for a short time highly infectious and his non-immune companions, if infected, promptly develop influenza.

Before discussing the evidence for the new concept it seems advisable to attempt a brief review of the rapidly expanding knowledge concerning the modes of parasitism of the influenza viruses in the animal kingdom and of their behaviour

in cell and cell cultures. Such a conspectus should provide a viewpoint of the various host parasite relationships that have been achieved by these viruses from which to judge the reasonableness of the speculations to be discussed.

Protracted symptomless infection, latency of whatever type, though well recognized as a characteristic of other virus groups such as herpes viruses, had been supposed until recently not to be a feature of orthomyxovirus infections. It has now been found that persistent infection with influenza A virus and latency of its subviral residues occur not uncommonly in nature and can be induced in cell cultures. The next section gives an account of modes of host parasite relationship of influenza A virus with non-human host species occurring naturally or in the laboratory, and of relationships at the cellular level.

The classification of human influenza A viruses has undergone a series of changes since their discovery in 1933, and the latest nomenclature has not met with general approval. Classification depends in large measure on epidemiological findings and, conversely, it may itself shape our epidemiological concepts. The matter is therefore considered later (pp. 13 17 ff.) in relation to the apparent evolutionary history of the influenza virus and its present degree of adaptation to various hosts, particularly to man. The latest nomenclature causes awkwardness in indicating the identity of some influenza A viruses of man, and when necessary the previous classification has been used thus: H1N1 (old style). The term major serotype is used in this paper for any major variant of influenza A virus whether caused by mutation or by genetic reassortment.

A subsequent section (pp. 17 22) attempts to assess how far along the road to speciation the human influenza A and B viruses have evolved, and to describe the natural history of the parasitic relationship with man.

Then there is (page 22 ff.) a consideration of the intriguing problems of the origins and successive prevalences and absences of major serotypes of human type A influenza virus. The current 'measles model' is powerless to explain these phenomena. Various speculations and the relative merits of anthroponotic and zoonotic hypotheses are discussed.

The new concept provides relatively simple explanations for aspects of the behaviour of influenza A that are inexplicable by the concept of direct spread. The evidence, previously presented in a series of papers (Hope-Simpson, 1979, 1981, 1983, 1984, 1986) is then reconsidered (pp. 31 40) prior to a summary of the conclusions of the authors.

MODES OF PARASITISM BY INFLUENZA A VIRUSES

Multiplicity of modes of parasitism

For a long time acute influenza and transient subclinical infections in man, the pig and the horse were thought to be the only modes of parasitism open to influenza virus. The infections were thought to be self-limiting, the virus being eliminated within a week or two having provoked only a transitory immunity in the host.

Since the discovery of avian influenza it has become apparent how varied and complex are the modes of parasitism that have been evolved by influenza A viruses in numerous host species. Many circumstances help to determine the host parasite relationship. The species and the age, sex and nutritional state of the host are

clearly important, as are the presence of other parasites or diseases, the exact serotype of the influenza virus, the previous influenzal experience of the host, and the geographical location and social interactions of the host species. Although there are dangers in transferring knowledge concerning the modes of parasitism in other host species, it is prudent to be aware of the many host–parasite relationships now known to have been adopted by influenza A viruses when we attempt to understand the behaviour of the virus in relation to mankind.

Influenza A virus parasitism in natural infection of non-human hosts

The non-human species, domestic and wild, now known to experience influenza A virus infections are so numerous that only a few can be mentioned that exhibit various modes of parasitism.

Since 1918, swine have suffered periodic outbreaks of influenza caused by Hsw1N1 (old style) influenza A virus first in the USA and later throughout the world. The Hsw1N1 virus is thought to be the naturally swine-adapted homologue of the virus that caused the great human influenza pandemic in 1918 (Laidlaw, 1935; Shope, 1958). In addition to suffering attacks of acute influenza, swine can harbour latent Hsw1N1 influenza A virus (Wallace, 1977, 1979), and breeder sows can act as carriers of latent Hsw1N1 strains and transmit them to their offspring (Mensik, 1962; Nakamura *et al.* 1972). Both Hsw1N1 and H3N2 strains have been isolated from naturally aborted pig foetuses and from stillborn piglets of sows that had been naturally infected in seasonal epizootics (Gourreau *et al.* 1985), thus confirming the early suppositions of Young & Underdahl (1949) that virus from infected pregnant sows can be transmitted across the placenta to cause abortion and stillbirth. Transplacental transmission does not necessarily cause death of the conceptus but may lead to symptomless latent infection of the offspring (Mensik, 1962). Epizootics of acute influenza in herds of swine latently infected with Hsw1N1 virus may be being precipitated by an unidentified stimulus linked with seasonal changes (Easterday, 1975).

Many species of bird harbour influenza A viruses, and ducks, like swine, may be of peculiar importance for human influenza because of the global distribution of both domestic and wild ducks, their high harbourage of varied influenza A viruses, the migration patterns of wild ducks, and the close association of domestic ducks with mankind in some communities. As in many other avian hosts, the viral association with the duck is usually commensal rather than pathogenic or latent. Healthy domestic and wild ducks may harbour large populations of varied influenza A viruses in alimentary canal and bursa. The virus continues to replicate without causing illness, especially in ducklings and young ducks, although at first there is a transient rise of haemagglutination-inhibiting antibody (Sinnecker, Sinnecker & Zilske, 1982). No rise occurs in specific neutralizing antibody (Slemons & Easterday, 1978). The faeces of ducks older than 70 days are often free from virus. A natural mode of parasitism seems to be a cycle in which infected lake water and faecal shore contamination by juvenile birds renew the parasitism in flocks of wild duck at their annual congregation in the breeding season. The process requires recurrent introduction of ducklings on to contaminated water (Hinshaw, Webster & Turner, 1978, 1979; Markwell & Shortridge, 1983).

The tolerance of ducks for influenza A virus explains the large number of major serotypes found in this host species, and the absence of specific antibody may

explain the finding that the same bird may often harbour several major serotypes simultaneously. All the 30 known combinations of genes coding for major HAs and NAs have been found in ducks (Alexander, 1982). The importance of this huge natural reservoir of influenza A virus genetic material in relation to the origin of novel major serotypes of human influenza A virus is considered later.

Turkeys often suffer severe epizootics of influenza A (fowl plague) with high mortality and protracted illness of survivors. The virus may, however, become latent in turkeys (Robinson, 1975).

In chickens fowl plague may also cause heavy mortality ranging from 40 to 100%, but in this species mild epizootics also occur. Much light has been shed on the problem of pathogenicity by a study of the disease in chicken flocks in Pennsylvania. A mild influenza A outbreak occurred in April 1983 caused by H5N2 strains, but in the following October the same virus suddenly began killing large numbers of poultry. Chickens infected with the virulent strain shed much virus in their droppings, and it was also isolated from their eggs and from houseflies in the chicken houses. The dead birds had suffered a general infection of all organs. The virulent strain was found to have been derived from the avirulent strain by a selective adaptation involving loss of the defective-interfering particles (DIPs) of virus present in the avirulent strain. This well-researched incident emphasizes the valuable role probably played by DIPs in mitigating the severity of influenzal infection (Bean *et al.* 1985). The acquisition of virulence was associated with changes at amino acid residue 23 near the stalk of the haemagglutinin and at amino acid residue 78 near antigenic site E (Webster, Kawaoka & Bean, 1986).

Situations inevitably arise in which individuals of one species are exposed to the influenza A virus parasitic on another host species, and interspecies cross-infection occasionally occurs. The opportunity must be greatest for aquatic species. In the 1979–80 season some 20% of harbour seals (*Phoca vitulina*) on the north-east coast of the USA died of pneumonia caused by an avian influenza A virus closely related to that of fowl plague, a virus not previously found in mammals. Another avian strain previously found only in birds killed harbour seals off New England in 1982–3. Unlike any previous mammalian isolate, both these seal viruses replicated like avian viruses in the gut of chickens, so avian viruses appear to have caused natural outbreaks of influenza in seals (Webster *et al.* 1984). Swine influenza has been reported as spreading in turkey flocks (Andral *et al.* 1985).

Haemagglutinins analogous to human H1, H2 and H3 have been found in water birds, crows, chickens, bats, whales, squirrels, deer and lake water (Lvov & Zhdanov, 1983).

The natural modes of parasitism of influenza A virus in non-human hosts therefore include at least the following: acute infection with and without illness, persistence, commensalism, vertical (transplacental) transmission and a variety of interspecies transmissions.

Experimental infection of non-human hosts

The ferret was the first animal found to be susceptible to artificial infection by human influenza A virus, and the ferret-adapted strains were found to be able to infect mice. Mice could also be directly infected with virus of human origin by several inoculation routes and by the respiratory passages after ether inhalation. Acute pulmonary infection in mice causes inflammation in the lungs, and although

infectious virus disappears after 9 days, viral antigen persists in the alveolar cells in high concentration for more than a year (Jakab, Astry & Warr, 1983). Protracted infection in the mouse leads to antigenic changes in the virus. An H3N2 strain, A/Hong Kong/1/68, could be isolated from mice for 45 days and periodically thereafter for 9 months, but after 5 months the strain showed changes (Frolov, Shcherbinskaya & Gavrilov, 1981). After 2 months infection with an H3N2 strain, the mouse spleens were found to yield H0N1 virions (Frolov *et al.* 1978). After 7 months, pathogenicity decreased or was lost and the viruses isolated between 2 and 8 months differed from the original strain in thermal stability (Frolov, Shcherbinskaya & Sklyanskaya, 1981).

The duration of infection is found to vary with the type of challenge. After live vaccine latency in lung tissues lasted for 35 days, whereas after disease it lasted 112 days. Persistent virus was found in the blood and viscera of offspring of mouse dams who were carriers of latent influenza A virus (Zuev *et al.* 1981). Such offspring were infected transplacentally whether the dams had been infected before or during the pregnancy. Infectious virus was found in the blood, lungs, liver, kidneys, spleen and brain of the baby mice, and survivors developed a 'slow virus' illness with stunting of growth and involvement of hypothalamus, immunocompetent organs and endocrine systems, and they ultimately died of the disease (Zuev, Mirchink & Kharitonova, 1983).

Whole virus persisted for only 30 days in chickens carrying influenza A virus, whereas HA and NA antigens persisted for 60 days. Hydrocortisone administered on day 50 provoked the appearance of several strains unrelated to the original virus (Smolensky *et al.* 1978).

The carrier state induced in turkeys could not be reactivated by heat, cold, thirst or hunger, but crowded transport provoked a prolonged elevation of H1 antibody (Robinson, Easterday & Tumova, 1979).

Such experiments have added the following information on the modes of parasitism available to influenza A virus: antigenic changes, some of them major, may take place during protracted infection; a slow virus type of parasitism may result from transplacental transmission in mice. The viral antigen may persist in the lung parenchyma of mice and chickens after infectious virus has disappeared, and infectious virions may be recalled during this period by the administration of hydrocortisone. These findings have conceptual importance on our attempts to understand the epidemiology of influenza, suggesting possible explanations of the intractable problems of the human disease. It is therefore important that the findings described above be confirmed in other laboratories to establish them on a firmer basis.

A distinction needs to be made between persistence of virions and of viral antigens, the latter usually succeeding but occasionally preceding the former. Persistent infection seems to provide the best conditions for promoting variation of the virus. The word latency may need to be restricted to the persistence of subviral residues, retaining the term persistent infection for protracted infection with some sort of virion production. Chronic infection describes persistence with regular (even if scanty) production of infectious virions (Medvedeva, Aron & Golubev, 1984*a*).

Protracted infection of cell cultures with influenza A virus

The type of cell in cell cultures, like the host species in animal infections, influences the result of infection by influenza A virus (Ahmed *et al.* 1981). Indeed, the host cell may select variants of influenza A virus and may be exerting as much evolutionary selective pressure as antibody (Schild *et al.* 1983); Nohinek, Gerhard & Schulze, 1985; Patterson & Oxford, 1986).

DIPs of influenza A virus are needed to initiate persistent infection, and its maintenance may require continuous production of temperature-sensitive (ts^-) mutants (Azadova, 1980; De & Nayak, 1980; Medvedeva, Aron & Golubev, 1984*b*; Medvedeva *et al.* 1985). Interferon may play a role but is not essential. During a persistent infection of 158 days, ts clones rose from nil to 100% by day 62 (Aron, Medvedeva & Golubev, 1981). The role of DIPs is considered in more detail on page 21.

Changes in the virus occur more rapidly during persistent infection than during production of standard infectious virus. Rapid and continuous evolution occurs in the DIPs, whereas acute infections with standard virus do not much alter the oligonucleotide map (Holland *et al.* 1979). During p.i. in human embryo kidney (HEK) and lung (HEL) cell cultures for periods from 40 to 289 days, 102 influenza A (H3N2) viruses were isolated. Of 44 of them examined 31 had preserved the original antigenic profile of haemagglutinin and neuraminidase, whereas it was markedly and permanently changed in 13 of them (Golubev & Medvedeva, 1978).

During a persistent infection of MDCK cells for 165 days with A/Victoria/35/72 strain of H3N2 influenza virus visible signs of virus reproduction were lacking, only 0·02–0·05% of the cells contained the influenza virus antigen, and virus isolation was irregular, and only possible with special methods. Interferon and DIPs were found not to have been responsible for the persistent infection in this system (Medvedeva, Aron & Golubev, 1984*a*). From day 9 there was progressive selection of ts mutants, and the inhibitor sensitivity increased and plaque phenotype of the persistent virus changed until at 158 days the population consisted entirely of temperature-sensitive, inhibitor-resistant small plaque clones. The haemagglutination-inhibition (H1) test showed no antigenic change in the haemagglutinin of the persistent virus from that of the original virus (Medvedeva, Aron & Golubev, 1984*b*).

The thermolability of the haemagglutinin and neuraminidase of these late mutants was increased, and the electrophoretic mobility of the H2 polypeptide was altered as its molecular weight decreased. Multiple ts mutations appeared in the genes coding for P2, NP, NA, M and NS proteins (Medvedeva *et al.* 1985).

In an H0N1/RES cell system maintained from 126 to 146 days, cycles occurred in which cytopathogenicity (CPE) alternated with cell regeneration. The virus from the cell-culture fluid from day 27 to day 105 would replicate in low multiplicity in chick embryo without producing haemagglutination. In one of five cell lines haemagglutination returned spontaneously during the last days of survival of the culture, and in two other lines it returned after serial passage in eggs and in PS cells. The other biological properties that had been lost also returned with

https://doi.org/10.1017/S0950268800066851 Published online by Cambridge University Press

the haemagglutination (Gavrilov *et al.* 1972). Defective interfering particles alternate with the standard virions (that cause cytopathic effects) in recurrent cycles for a time, but thereafter DI particles alone are produced (Kantorovich-Prokudina *et al.* 1980). Periods of quiescence may then occur in which the virus seems to have been lost, but later haemagglutination and other activities may return spontaneously, demonstrating that the viral RNA had not been lost but had been present in some mode of latency (De & Nayak, 1980).

Both past and future antigenic variants have been found to evolve during p.i. in cell culture. For example, A/Hong Kong/1/68 (H3N2) virus produced both A/Victoria/3/72 (H3N2) and A/Singapore/1/57 (H2N2). The haemagglutinin and the neuraminidase had changed both forwards and backwards (Golubev, 1975). The finding of spontaneous antigenic shift in cell culture has not yet been repeated in other laboratories. Recent techniques detect previously unsuspected viral residues. Thus after paramyxovirus infection in mice, when all other evidence of viral presence had disappeared, the viral RNA was present life-long in brain tissue, detectable by RNA hybridization with a cloned genomic DNA probe. The latent viral RNA expressed no protein (Koch, Neubert & Hofschneider, 1984). Similar techniques have shown synthesis of influenza A virus RNA in persistent infection in MDCK cells, and also demonstrated cRNA and vRNA synthesis during the latent phase when there was no cytopathic effect and no virus could be isolated (Stelmakh, Medvedeva & Golubev, 1982).

Infectious virus accumulates in cell lysosomes during chronic infection, and the possible existence of a lysosomal carrier state has been proposed. Acid phosphatase, present in acute infection, is absent in chronic infection (Maslovsky, Neklyvdova & Orlova, 1979). The nature of the complex containing both polysomes and vRNA is not known, but it is not caused by contamination with vRNP (Nayak, D'Andrea & Wettstein, 1976).

Influenza A virus, then, shows a variety of modes of behaviour even in such a relatively simple host–parasite environment as cell culture: acute cytopathogenic productive infection; persistent infection with recurrent cycles of loss of cytopathic effect and other properties; latency with no production of virions; spontaneous reactivations from latency and production of variants during persistent infection with opportunity for antigenic drift and perhaps antigenic shift, particularly if the particles encounter immunity specific for the parent strain. The observations of Medvedeva, Aron & Golubev (1984*b*) indicated that selection of ts^- mutants during persistent infection was not fortuitous, but a regular process leading progressively to complete displacement of variants with ts^+ phenotype from the viral population.

Immune pressure on laboratory culture of influenza A viruses has long been used to select variants (Archetti & Horsfall, 1950; Isaacs, 1951), and attempts have been made to simulate natural antigenic drift and to anticipate the natural evolution of novel variants (St. Groth & Hannoun, 1973; St. Groth, 1977). Such experiments have provided laboratory support for the new concept of epidemic process in relation to antigenic drift as discussed later.

The rate of drift (sequence change) in the gene coding for H is similar to that in antigenically unrelated genes, and it does not tend towards any other major serotype. It has been possible to construct a dendrogram of the probable evol-

utionary development of all the known major HA serotypes from a presumed common ancestor (Air, 1981).

EVOLUTION AND THE SYSTEMATICS OF HUMAN INFLUENZA A VIRUSES

Speciation

The speciation of living creatures has been produced by selective environmental pressures upon their random variability, eliminating variants less suited to the particular environmental niche and favouring more suitable variants. The resulting adaptations have ultimately divided the varieties into different species, and the process is facilitated by natural barriers that isolate creatures within a particular environment and prevent genetic mixing with related creatures from other areas. Mountain ranges, oceans and climatic rigours have provided barriers that have accelerated the production of many species of animal and plant.

For parasites the natural barriers corresponding to oceans and mountains are those very adaptive changes that have enabled them to survive in a particular host species and to achieve transmission from host to host within that species. Several peculiarities encourage rapid speciation of parasites. Adaptation is mutual, the host also developing adaptive changes (Ahmed *et al.* 1981; Nohinek, Gerhard & Schulze, 1985), and many parasites have developed a very high rate of reproduction that favours variation. Lacking almost every function except programming ability, viruses are amongst the most specialized of parasites.

A species has been defined as the totality of living creatures of a particular sort throughout many generations which inhabit a definite space, which are able to cross freely with one another, and which are separated from similar neighbouring totalities by isolation that may be spatial, seasonal, physiological and/or genetic (adapted from Timofeev-Resovsky, 1982). Human influenza A strains are not entirely insulated genetically from those of non-human hosts, but eco-physiological and spatial isolation provide barriers. Parasitism in different host species imposes specific adaptation, and different potentialities are developed by the virus in different sorts of host. Humans are seldom naturally infected with strains derived directly from animals and birds.

Classification and nomenclature

The global picture of influenza viruses as parasites of the animal kingdom is so complex that it causes difficulty in establishing an acceptable classification and nomenclature. The names of the influenza viruses parasitic on man have undergone a rapid series of changes as knowledge has grown, and the latest nomenclature has not met with universal approval. Influenza A and B viruses share sufficient genetic material to bespeak a not-too-remote common ancestry and so may be assigned to the same genus (Skehel & Waterfield, 1975; Scholtissek, Rohde & Harms, 1977; Alexander *et al.* 1981; Krystal *et al.* 1982). They differ sufficiently to belong to separate species. Influenza B virus, a human parasite, is infrequently transmitted to non-human hosts (Chang *et al.* 1976), and genetic reassortment between influenza A and B viruses occurs rarely and seems not to have produced a successful hybrid (see Tobita & Ohori, 1977). Influenza B virus lacks subtypes such as characterize human influenza A virus (Chakraverty, 1971). Thus influenza B virus

seems already to have evolved into a separate species during its human parasitism (Alexander *et al.* 1981).

The situation with influenza A virus is not so straightforward, and it seems desirable for the present to retain all strains from whatever host within a single species. Within that species are numerous varieties, some being major and some minor variants.

Speciation may be gradual, with no clear division of varieties from species. Influenza A viruses are sometimes transmitted from one sort of host to another, and hybridism by genetic reassortment between variants occurs readily in the natural setting (Webster, Hinshaw & Bean, 1977; Desselberger *et al.* 1978; Yamane *et al.* 1978; Bean, Cox & Kendal, 1980). Nevertheless, one should not underestimate the degree to which some variants of influenza A virus have become specifically adapted to particular host species (e.g. Scholtissek & v. Hoyningen-Huehne, 1980) and, where the same influenza A virus has been found in more than one sort of host, the modes of parasitism have usually differed. For example, the virus that is commensal in sea birds may cause lethal pneumonia in seals, and H3N2 strains show a behaviour in swine different from that in man.

Classification of human influenza A viruses

The influenza A viruses parasitic on mankind exhibit a singular behaviour differing from that yet found in any other host species. Each of five major serotypes has had successive eras of many years of prevalence during which, having achieved worldwide distribution within the first season, it has usually continued to cause almost all the human type A influenza in a series of epidemics for the rest of its era, except since 1977 and possibly also from 1907 to 1918, when two major serotypes have co-circulated. The pattern of repeated sequences of world domination with long intervals separating the reappearance of the major serotype suggests an evolutionary adaptation whereby the influenza A virus has secured its survival as a human parasite, and the phenomenon should somehow be reflected in the virus nomenclature. The latest nomenclature approved by the WHO Experts' Committee in February 1980 fails to do so because it no longer recognizes the contribution of the host species. Based on the double immunodiffusion test, it records valuable information about relatedness of influenza A strains regardless of host species, but conceals stable differences of biological and practical importance recorded by the haemagglutination-inhibition test, on which an earlier nomenclature was based.

Soon after the discovery of human influenza A virus it was noticed that viruses isolated in the same epidemic season were much alike, whereas those from different seasons differed antigenically, though the differences were not great, and influenza caused by an earlier strain protected against reinfection in later seasons. Strains isolated before 1946 were later considered to have belonged to a single major serotype whose minor variants were distinguished by this cross-protection, and the group was awarded subtype status, designated H0N1. Vaccination with any H0N1 strain conferred some protection against all the minor variants of the H0N1 major serotype.

In 1946 a novel major serotype, 'A prime', made its epidemic appearance, later designated H1N1 because its haemagglutinin differed from that in the H0N1 (old

style) strains. The H0N1 (old style) viruses promptly disappeared, and H1N1 (old style) strains caused almost all the influenza A in the world population for the next 11 years, and previous infection or vaccination with H0N1 strains afforded no significant protection against them (Francis, Davenport & Hennessy, 1953). H1N1 therefore also became a subtype designation.

World dominance by H1N1 strains ended abruptly in 1957 when they were replaced worldwide in the pandemic caused by H2N2 influenza A viruses, a new subtype in which both surface antigens had changed. H2N2 viruses also had an era of 11 years of solo world prevalence.

The era of H2N2 influenza A viruses ended in 1968 with their replacement worldwide by H3N2 strains in which only the haemagglutinin had changed significantly. The subtype of H3N2 viruses had near-solo dominance until 1977.

These experiences led to a belief that during their era of prevalence strains belonging to one subtype somehow excluded strains belonging to all other subtypes of influenza A virus, though tolerating the presence of influenza B viruses. This apparent rule was broken in 1977 when H1N1 (old style) viruses reappeared after 20 years' absence, rapidly achieved world-wide distribution, and are still in 1986 co-circulating with H3N2 viruses.

The classification into these four separate subtypes of human influenza A virus – H0N1, H1N1 (old style), H2N2 and H3N2 – was agreed by a committee of experts meeting at Atlanta, Georgia, USA, in November 1978 (W.H.O., 1979), and a fifth subtype, Hsw1N1 was added, representing the viruses thought to have caused all the human type A influenza from the 1918 pandemic until 1929, when they seem to have been replaced by H0N1 strains. Pigs, it is supposed, became infected from humans in 1918, and these human-derived Hsw1N1 strains then became adapted to survival in swine (Laidlaw, 1935; Shope, 1936; Langmuir & Schoenbaum, 1976).

The term subtype in the 1978 classification indicated a group of related influenza A viruses that had had an era of a number of years of world prevalence in man, often initiated by a pandemic. Successive epidemics in each subtype era were noticed to become progressively less severe. Viruses of the same subtype protected against one another by natural infection or by vaccination much more strongly than against strains belonging to other subtypes. The antigenic shift that inaugurated each era of prevalence by a new subtype was thought to differ in kind from the point-mutations in the genes coding for the haemagglutinin and the neuraminidase that cause antigenic drift within each subtype – shift was thought not to be simply a large drift. This assumption was challenged by the finding that, whereas the changes from H1N1 to H2N2 and from H2N2 to H3N2 were caused by genetic reassortment, a sort of hybridism (Scholtissek *et al.* 1978), the change from H0N1 to H1N1 was caused by mutation in the gene coding for the haemagglutinin differing in degree though not in kind from those causing antigenic drift (Scholtissek *et al.* 1977; Air, 1981).

Two causes of major antigenic change, mutation and reassortment, had thus been found to have produced similar consequences in the relationship of influenza A virus with the human host species. The subtype classification might have been modified to indicate the sort of change, but about the same time in non-human host species influenza A viruses were being found with genes similar to those coding

for the haemagglutinin and the neuraminidase in human strains (Zakstel'skaya *et al*, 1980), a finding that seemed to necessitate a classification that was independent of the host species (W.H.O., 1979).

The double immunodiffusion test showed similarities in the haemagglutinin of Hsw1N1, H0N1 and H1N1 (old style) so that these three major serotypes were amalgamated into the new subtype H1N1 (Zakstel'skaya *et al*. 1980), reducing the number of human influenza A subtypes from five to three, and concealing the major change in the human-host/influenza A parasite relationship that had occurred about 1929 and again in 1946. The new classification also conceals the phenomenon that the three amalgamated subtypes had each had its era of many years of solo prevalence as the dominant influenza A virus of mankind, during which each had developed its own family of mutually protective minor variants, and that immunity conferred by viruses of any one subtype was largely ineffectual against strains of the other two. Furthermore, each possessed its own constant profile of inhibitor sensitivity (Smorodintsev *et al*. 1981*a*, table 2). The unification of the three subtypes as H1N1 impedes our understanding of the natural behaviour of influenza A viruses in mankind and possibly also in other host species, and it raises difficulties for vaccine prophylaxis. There are thus good reasons for recognizing the original five serotypes in the nomenclature of human influenza A viruses.

According to the doctrine of original antigenic sin 'the prevalence and magnitude of serum antibody against each individual influenza A strain are greatest in those who were young, and therefore probably experiencing their first attack of influenza, when the strain was circulating in the world and causing human epidemics' (Francis, 1960). Persons having suffered an H0N1 influenza A virus infection who were subsequently vaccinated with H1N1 (old style) vaccine were found to produce an antibody response against H0N1 exceeding that against the H1N1 vaccine strain. A problem in classification arose when it was later found that such anamnestic reactions of H0N1 and H1N1 antibodies are not provoked by H2N2 virus infections. In relation to original antigenic sin there thus appeared to be two classes of influenza A viruses, one containing Hsw1N1, H0N1 and H1N1 strains and the other containing H2N2 and H3N2 strains (Marine & Thomas, 1979). Not all observers, however, agree that H3N2 viruses fail to stimulate anamnestic antibody production in persons previously infected with Hsw1N1 influenza A virus (Masurel, 1976). The breaching of the rule that successive major serotypes have eras of solo world dominance in 1977 when H1N1 strains returned worldwide during an era of H3N2 prevalence suggested that solo dominance may operate only within each of the two classes as defined by original antigenic sin. H1N1 strains may attack persons very recently infected with H3N2 virus (Frank, Taber & Wells, 1983).

The role of the neuraminidase antigen may have been undervalued (Naikhin *et al*. 1983). Neuraminidase varies independently of HA, and a new nomenclature of human influenza A viruses has been proposed restoring the status of Hsw1, H0 and H1 (old style) and identifying the three variant neuraminidases in both N1 and N2. Influenza A and B virus are therein classified as separate species in the family of orthomyxoviridae, the five old-style subtypes as serotypes, and the main antigenic variants of each serotype as serosubtypes (Smorodintsev, Golubev & Luzjanina, 1982, table 4).

Influenza C virus appears to have a common ancestry with influenza A and B viruses and must therefore be supposed to be the earliest of the three to become a human parasite. It has established a more harmonious relationship with its human hosts, seldom causing epidemics and frequently causing no disease at all.

NATURAL HISTORY OF THE MAJOR SEROTYPES OF HUMAN INFLUENZA A VIRUS

Origins of the major serotypes

Historical considerations

Two aspects of the origin of major serotypes of human influenza A viruses need to be distinguished. First, how and whence did mankind originally acquire each major serotype? The H0N1 strains may have originated from a mutation in the gene coding for the haemagglutinin of Hsw1N1, and the H1N1 strains may have originated similarly by a mutation in the gene coding for the haemagglutinin of H0N1. We do not yet understand the initial origins of Hsw1N1, H2N2 and H3N2. Secondly, how and whence does mankind continue to acquire these viruses for recurrent cycling? They have achieved worldwide prevalence in a single season each time they have reappeared during the last half-century. Each time the previous major serotype, that had caused virtually all the human influenza A in the world for a decade or more, abruptly disappeared and next season was replaced worldwide by viruses belonging to the next major serotype. One cannot overemphasize the importance of this phenomenon in the construction of a valid epidemiological hypothesis. The fact that a similar phenomenon occurs during antigenic drift suggests that antigenic shift and drift possess a similar mechanism.

Hypotheses

Kilbourne has proposed a model that has much to commend it as an explanation of recurrent origins of major serotypes in the human population (Kilbourne, 1975). Each major serotype is presumed to be transmitted from man to some non-human host species that, like the pig in some parts of the world, lives in close association with humans. In the alternative host the virus remains antigenically stable as an enzootic parasite and, although frequently transmitted back to human associates, it usually fails to regain human prevalence because of an unfavourable immune situation in the population. Sooner or later enough persons will have died and enough been born to form a largely non-immune population, and the virus transmitted back from the alternative host species seizes the opportunity to establish another era of prevalence in the world population. Within a dozen years or so it will again have exhausted the supply of non-immune humans and would become extinct, but for the enzootic infection in the alternative host species patiently maintaining the virus strain until the next gap in human population immunity provides the conditions for yet another era of prevalence in mankind.

Here we also have a mechanism whereby novel major serotypes may arise. Influenza A viruses specific to the alternative host, and those received by it from other non-human host species, may encounter the human strains, and genetic reassortment may occur, thus providing a route whereby novel genes can gain access to parasitism in the human population (Andral *et al.* 1985). Such hybridism

has been providing in nature a mechanism for the evolution of influenza A virus as a parasite of many host species possibly including man, and appears to continue in operation (Lvov *et al.* 1978).

Kilbourne's attractive hypothesis gains credibility by the natural behaviour of Hsw1N1 and H3N2 influenza A viruses in swine and mankind (Shortridge, Cherry & Kendal, 1979; Ottis *et al.* 1982; Masurel *et al.* 1983). Valuable as the model is as possibly explaining the first origin of some major serotypes of influenza A virus in mankind, it does not explain the current behaviour of human influenza A. The pig or other host would need to be co-ordinating transmissions to man worldwide for each successive major serotype to appear worldwide within its first season. Human case-to-case spread from a single porcine source could not encompass the world population at such speed. Herald waves of influenza caused by the epidemic strain in the pre-epidemic season (Marine, McGowan & Thomas, 1976; Glezen, Couch & Six, 1982) only shift the problem back a year because the pre-epidemic wave would itself need to have been worldwide. The model also fails to explain the abrupt disappearance of the predecessor serotype, and of other features such as the ability to cause a large final epidemic as in January–April 1968, when most people had already been immunized by seven previous epidemics in the H2N2 era. The indiscriminate age distribution of those attacked in the final epidemic is also unexplained (Hope-Simpson, 1984).

The Kilbourne model would be expected to produce a complicated pattern of simultaneous parasitism by varied influenza A viruses belonging to numerous major serotypes, rather than the ordered succession of clearly demarcated eras of prevalence, each consisting of a limited series of world epidemics from which only a few incommunicado communities altogether escape. Influenza A has survived in this way as a successful human parasite for at least a century, possibly for many centuries.

Parasitic strategy

Epidemiological findings have elucidated the requirements of the influenza A virus for a parasitic strategy within the human host species. The virus needed to evolve some adaptation whereby it could limit the proportion of the population attacked during each epidemic. A clue as to how such limitation is being achieved can be found in the wide age distribution of the persons attacked in each successive epidemic. The new concept in this paper proposes a simple mechanism by which this is achieved (see page 38).

A major serotype infects and immunizes almost all susceptible members of the world population during each era of its prevalence (Gill & Murphy, 1985), so that it cannot again become prevalent until sufficient non-immune subjects have been recruited by births. The approximate intervals separating successive major serotype eras were: for H2N2 strains (1900–57) 56 years; for H3N2 strains (?1917–68) 50 years; for H1N1 old-style strains (?1917–46) 28 years and also 1956–77, 20 years. Influenza A viruses must therefore have evolved adaptations for retaining each major serotype relatively unchanged during these long intervals, and for reactivating and recycling them when a suitable gap in population immunity offered the opportunity for an era of renewed world prevalence. The viral and serological anachronisms found in the human population between their eras of

prevalence indicate that the adaptations for retention and recycling may be operating within the human host parasite relationship (Belyakov *et al.* 1983 and also page 27).

The changes that occur in the virus during these long retentions between eras of prevalence are remarkably small. Both the paucity of changes and the changes that do occur may illuminate the adaptive mechanisms. After a quarter of a century's absence H1N1 strains reappeared as a reassortment containing one gene of the human prototype strain of 1947 and the other seven genes of a minor variant that had caused a large epidemic in 1951 (Kendal *et al.* 1978; Shilov *et al.* 1981). Later work has decided that the 1977 strain of H1N1 virus is identical in all its genes to virus A/FW/50/H1N1 of the earlier H1N1 era (Golubev, 1984). The 1977 strain had received no contribution from a non-human influenza virus between 1950 and 1977 (Nakajima, Desselberger & Palese, 1978).

Serological studies in elderly persons indicate that the neuraminidase of H3N2 viruses of the previous era of prevalence (*c.* 1900–17) differs from that of the most recent era since 1968, and that the neuraminidase of the still earlier H2N2 strains of the nineteenth century era resembled that of equine 2 influenza A virus. The finding prompted Webster & Laver (1975) to comment: '...recycling of influenza viruses may result from emergence of viruses from an animal reservoir, either with or without a recombinational event, when herd immunity no longer precludes it from the human population'. Here again, detailed consideration of what must actually have taken place is difficult to reconcile with the suggested interspecies transmission. Transfer from horse to man in a single locality could not have resulted in the appearance of the hybrid virus in all parts of the globe within one season by direct spread from the sick. Moreover, such interspecies transmissions would be expected to produce a simultaneous assortment of major serotypes in mankind as it has in birds. We await the next antigenic shift to throw more light on the question of interspecies transfer of genetic material to humans from non-human hosts.

Genetic similarities between influenza A viruses infecting different host species are not easy to interpret. They speak of evolutionary relatedness, and some may result from a recent hybridism perhaps involving a major human serotype (Nakajima *et al.* 1982). Similarities of evolutionary interest may, however, bear little relevance to current epidemic behaviour of influenza A. Influenza virus appears to be in the process of evolving a specific pattern of parasitism adapted to each species of host, that of influenza B virus as a parasite of man being an example. Influenza A virus is still capable of varied modes of parasitism in many host species. It seems to be developing a more complicated relationship with mankind in which the mutual adaptations of host and parasite have not yet so specialized the virus as to insulate it from being transmitted to other host species and from hybridizing with strains that are also in the process of becoming adapted to parasitism in other hosts. The human respiratory parenchymatous cells seem already to have evolved considerably less resistance to infection by human influenza A viruses than to those from other hosts. Thus speciation of influenza A virus seems not yet to have been completed either in man or in any other host species.

Nevertheless, in man influenza A virus has gone some way towards speciation.

Host range may be determined by a single gene (Almond, 1977; Buckley-White, Naeve & Murphy, 1986) and different host cells determine different subpopulations of the virus (Schild *et al.* 1983; Patterson & Oxford, 1986). The haemagglutinin of human H0N1 strains is closely related to that of human H2N2 strains (Davis *et al.* 1981), and the relation between the H1 (old style) and H2 haemagglutinins of human strains is at least as close as that between early and late H1 strains (Gengqi *et al.* 1980). The H2N2 viruses of ducks differ from the human H2N2 strains in oligonucleotides, avidity and inhibitor profile (Nerome *et al.* 1984). The H3 receptor on the haemagglutinin molecule seems to be specific for the species of host from which the virus was isolated (Rogers & Paulson, 1983).

Epidemic strains of influenza A virus are neither genetically nor antigenically uniform. When H1N1 and H3N2 strains are – as now – co-circulating in man, recombinants commonly occur, but they have no long-term advantage over non-recombinant strains. There seems to be a comparable selective pressure and fidelity of replication for each genotype. Genetic variants found by oligonucleotide mapping depend on time, with base sequences changing at 0·12–0·54% per annum for the haemagglutinin (Cox, Bai & Kendal, 1983). The non-structural genes evolve along a common lineage unaffected by reassortment, also diverging about 3·0% every 10 years (Krystal *et al.* 1983).

In 1976 for the first time a strain, Hsw1N1, indistinguishable from a non-human influenza A virus, was isolated from an outbreak of human influenza. Despite absence of antibody to Hsw1 and to N1 in much of the population, and total absence of these antibodies in persons under 25 years old, the virus failed to spread, although at that time H3N2 strains were spreading normally (Beare, Kendal & Craig, 1980).

Possible modes of protracted parasitism of man by influenza A virus

Requirements and possibilities

Anthroponotic derivation of recurrent major serotypes of influenza A virus in man presupposes persistent infection by the virus and/or latency of its residues in the human host tissues. What direct or indirect evidence supports such a conception? Most hypotheses of influenzal epidemiology tacitly assume that influenza A virus is surviving in mankind only by direct spread from the sick, an assumption that is untenable as the epidemic process operating in human influenza A, though possibly valid for other host species.

The new concept proposes that both persistent infection and latency occur as an integral part of the epidemic mechanism, preserving the virus between epidemics, and ensuring that it encounters the immune selective pressure that it has itself provoked in its host, so that antigenic drift occurs. If persistent infection is a regular property of the human/influenza A virus relationship there is reason to seek within the same relationship for the longer-term latency that would maintain the major serotype for many years between its cyclical eras of prevalence.

The requirements for persistent infection and latent human parasitism by influenza A virus are readily available. During acute influenzal illness the virus replicates within cells lining the respiratory passages, and the biosynthesis of haemagglutinin, its transport, glycosylization and cellular localization are all determined by functions of the host cell (Gething & Sambrook, 1981). Thus the host

may have a second mechanism at the cellular level for selecting virus variants before transmission (Schild *et al.* 1983; Patterson & Oxford, 1986). Early in the replicative process, before the synthesis of virus-specific RNA, host-cell DNA must be transcribed by DNA-dependent viral RNA-polymerase II (Spooner & Barry, 1977).

Defective interfering (DI) particles

DI particles, incomplete virions produced early in acute influenza, were at first considered abnormal, but it now seems that they may play an essential role in the evolution of the virus and in its adaptation to the host. A single particle suffices to inhibit the release of progeny virions from a cell infected with vesicular stomatitis virus (VSV), whereas two DI particles per cell interfere with this inhibition and the yield of standard virions is no longer totally suppressed. An avian influenza virus, the cause of fowl plague, behaves similarly (Carter & Mahy, 1982). Thus in low multiplicity DI particles can produce an all-or-none effect on viral productivity, and a single DI particle inhibits the formation by standard virus of an infectious centre in cell culture. Standard virions may be entirely replaced by DI particles within four growth cycles (Janda *et al.* 1979). A ratio of one between DI particles and standard virions reduced infectious yield by 99·9% without reducing the yield of DI particles. The interference by DI particles is most effective within the first 3 h and must therefore interfere with an early function of replication (Nayak *et al.* 1978). DI particles do not cause influenza, and their presence exerts a profound effect on influenzal illness. They moderate pathogenesis (Rabinowitz & Huprikar, 1979), and their absence appears to have permitted an exacerbation in epizootics of avian influenza causing a high mortality in chicken flocks (Bean *et al.* 1985).

Protracted infection and DI particles

We have considered DI particles in some detail because they are essential in initiating and perhaps also in maintaining persistent infection with influenza A virus (Huang & Baltimore, 1970; Bektimirov *et al.* 1976). Persistent infection encourages variation of the virus. After 1 year of persistent infection by VSV in BHK cells the infectious genome had evolved several changes in the oligonucleotide map, and after 3 years the changes were numerous, and continued to increase in subsequent years. Meanwhile repeated passage of standard virus in acute infections of several cell types *in vitro* or *in vivo* did not lead to oligonucleotide changes. The changes during post-infection were not confined to standard virions, the DI particles themselves also being altered. 'The sequestered intracellular environment of persistently infected cells favours rapid and continuous evolution'. Persistently infected cells 'survive indefinitely and allow a variety of virus mutants to arise and compete (and complement each other) for long periods without any need to mature and spread to other cells. Obviously the less virulent mutants would tend to be selected since the more virulent will result in cell death and be eliminated from surviving cell populations' (Holland *et al.* 1979). These observations favour the new concept of epidemic process proposed in this paper, and add to it the probability that the human case of persistent infection with influenza A virus will himself be exerting additional selective pressure on the

variants he is harbouring before he sheds virus to infect his non-immune companions.

Although DI particles do not themselves cause influenzal illness they contain all the genetic information required for the construction of standard infectious virions. During persistent infection there is co-evolution of host cell and virus, so that the host cell also plays a crucial role in maintaining persistent viral infection (Ahmed *et al.* 1981). Persistently infected cells undergo alternate cycles of cytopathogenicity and quiescence, ultimately succeeded by virus latency (Lacorte, 1974*a*, *b*; Koch, Neubert & Hofschneider, 1984). Latent subviral residues are not easy to discover (Stelmakh, Medvedeva & Golubev, 1982), and their presence may only be manifested when latently infected cells revert to production of virions (Perekrest *et al.* 1974; Johnson, Lazzarinen & Waksman, 1980; Zuev *et al.* 1981). Influenza A virus may remain persistent or perhaps latent as RNA–polysome complexes (Nayak, D'Andrea & Wettstein, 1976), or in a lysosomal carrier state (Maslovsky, Neklyvdova & Orlova, 1979). After 70 and 190 days passaging, viral proteins can be detected in cytoplasm and plasma membrane of the host cells by immune peroxidase techniques (Čiampor *et al.* 1981).

There can thus be no doubt that persistent infection and latency of influenza A viruses occur in nature. Latency of Hsw1N1 and H3N2 strains occurs naturally in swine. Persistent infection has been produced experimentally with avian influenza virus (Smolensky *et al.* 1978) and in mice (Frolov, Shcherbinskaya & Sklyanskaya, 1981), and in both species the unproductive infection led to changes in the virus. In mice the virus is not confined to the respiratory apparatus, for immune complexes consisting of antigen, antibody and complement can be found in kidneys and heart as well as lungs after virus challenge, after live vaccine or during convalescence from acute influenzal infection. The frequency with which the complexes are found depends on the level of serum antibody and the route of virus challenge. The complexes themselves may transmit the infection (Semkov & Wilczyński, 1979).

Of the three phases, acute viral infection secures an astronomical degree of replication, persistent infection secures rapid variation of the virus, and latency of subviral residues seems appropriate for maintaining the virus relatively unchanged for almost indefinite periods. A fourth phase, chronic infection with continuous low-multiplicity production of infectious virions, occurs in certain laboratory systems (Medvedeva, Aron & Golubev, 1984*a*) and presumably may occur naturally in some hosts.

THE ORIGIN OF MAJOR SEROTYPES OF HUMAN INFLUENZA VIRUS

Mutation versus reassortment and recombination

A major serotype – subtype in old terminology – designates a family of influenza viruses immunologically related through their H and N antigens, so that cross-protection occurs as between the different minor variants of any one major serotype. Historically each major serotype has had one or more periods of world prevalence in man, an era, alone or contemporaneously with another.

There is still uncertainty about how each major serotype has arisen. The novel strain might have been produced by a major mutation in the gene coding for the

haemagglutinin, or mankind might be obtaining novel strains as total virions or by genetic reassortment from the vast comprehensive genofond now known to exist in non-human hosts, particularly from domestic ducks and swine. Either process, mutation or hybridization, may have taken place long ago, and the then novel strains may have become adapted to the human host, perhaps over centuries, in cycles of latency and reactivation. The similarities of some genes of human influenza A virus strains with those of strains in non-human hosts might thus be providing evidence of the evolutionary history of the virus as a human parasite, and minor differences might indicate evolutionary divergence. The processes are not mutually exclusive and may all have been operating at different times and may still be at work.

Observations suggesting origins from non-human hosts

Interspecies transmissions

A zoonotic origin for novel human strains as whole virions or by hybridism presupposes the existence of the appropriate viruses in non-human host species, opportunity for transfer of the virus or its genetic material to man, and evidence that such interspecies transmissions to man have actually occurred and been succeeded by immediate adaptation of the novel strain to the human host. Moreover, the novel parasite must somehow have contrived to infect mankind worldwide within a few months and then to have sustained an era of general prevalence lasting for many years.

Most of these requirements are satisfied. Numerous non-human host species act as hosts to influenza A viruses belonging to many major serotypes. Domestic ducks and swine, both notable carriers, live in close association with each other and with man in rural areas of many parts of the world. All known combinations of the genes coding for haemagglutinin and neuraminidase have been found in domestic and wild ducks. Several pandemics appear to have originated in the People's Republic of China, and it has been suggested that a rural epicentre south of the Yangtse river may provide the genofond whence pandemics have been generated in the close association of the human population with their domestic creatures. The sluggish waters of ponds and rice paddies are suspected of maintaining and distributing the novel strains (Beveridge, 1977; Shortridge & Stuart-Harris, 1982; L'vov & Zhdanov, 1983; Laver, Webster & Chu, 1984).

In 1968, soon after H3N2 strains had first appeared in man in Hong Kong they were found infecting the local pigs, and a similar rapid transfer to swine has followed human H3N2 epidemics in many parts of the world, but the human infection has generally preceded the porcine. Minor variants of H3N2 virus subsequently arising from antigenic drift and causing successive human epidemics have also promptly appeared in swine in the areas affected (Popescu, Iftimovici & Jacobescu, 1976; Tumova *et al.* 1976; Chang *et al.* 1977; Wallace, 1979; Alexander *et al.* 1981; Arikawa *et al.* 1982; Ottis *et al.* 1982). H3N2 strains had not been found in swine before the first human epidemics of 1968, so the human strain was not acquired from the pig. Nevertheless, back transfer from swine to man does occasionally occur, so swine could be providing an accessible reservoir of old and newer strains for mankind (Masurel *et al.* 1983).

Less commonly H3N2 strains are naturally transmitted from man to other non-

human hosts (Popescu, Iftimovici & Jacobescu, 1976) including his closest associates such as dogs (Chang *et al.* 1976) and horses (Bronitki *et al.* 1974).

Of peculiar interest is the major serotype Hsw1N1, which supposedly caused the devastating human pandemic of 1918 and remained the sole cause of human influenza A until 1929. It is thought to have been transmitted in 1918 to domestic swine in the USA, where it became established as the natural cause of swine influenza, appearing in seasonal epizootics, and has since spread to swine in many other parts of the world (Shope, 1936; Wallace, 1977). In 1976 a reversal of the presumed interspecies transmission caused a sharp human outbreak of Hsw1N1 influenza A at Fort Dix, USA, in which a man died (Top & Russell, 1977; Davenport, 1977; Beare, Kendal & Craig, 1980). Not unreasonably the outbreak was feared to presage another pandemic like that of 1918 (Hattwick *et al.* 1976) but the virus failed to become generally prevalent. No direct contact with swine could be found to account for the Fort Dix outbreak, but contemporaneously a small Hsw1N1 outbreak occurred in which 3 of the 15 persons attacked had had direct contact with pigs (Thompson *et al.* 1976), and sporadic transmissions from pig to man were also reported at that time (e.g. O'Brien *et al.* 1977). In the Fort Dix outbreak only five isolates of Hsw1N1 virus were made 'despite extraordinary efforts' although co-circulating H3N2 strains were readily isolated (Goldfield *et al.* 1977). The Fort Dix outbreak evidently resulted from transmission from swine to man of Hsw1N1 virus that achieved only a limited adaptation to survival in the human host species (Beare, Kendal & Craig, 1980). Hsw1N1 seems to be a strain now well adapted to parasitism in swine, not uncommonly being transmitted to human hosts but not yet becoming re-established in the human host species for an era of prevalence as a dominant major serotype.

Hsw1N1 strains have been isolated from turkeys, and one of these avian strains may have caused influenza in a laboratory technician (Hinshaw *et al.* 1983).

Both H3N2 and Hsw1N1 viruses have remained genetically rather stable in swine, but are not always homogeneous even in the same individual pig or man. In 1976 the Hsw1N1 strains isolated from both hosts consisted of two populations with distinguishable haemagglutinins (Kendal, Noble & Dowdle, 1977). Two variants have been found, not only in pigs and man but also in birds, in 12 countries, a US type with the typical HA of A/New Jersey/8/76 (Hsw1N1) virus in Hong Kong, Japan and the USA, and a European type in Belgium, France, the Federal Republic of Germany and Spain with a distinguishable HA (Hinshaw *et al.* 1984).

Human influenza A strains can spread naturally to ducks (Tumova *et al.* 1975; Shortridge, 1979) in which the virus drifts antigenically more slowly than in the human host (Sriram *et al.* 1980; Maywald *et al.* 1982). Hsw1N1 strains from the duck can naturally infect swine (Hinshaw, Webster & Turner, 1978) and man (Scholtissek, Rohde & v. Hoyningen, 1978). Kaplan (1982) gives an excellent review of interspecies transmissions including those affecting man.

In summary, the natural genofond of influenza A virus genetic material in non-human hosts provides a reservoir from which transmissions to man can and do occasionally occur, most often with Hsw1N1 and H3N2 strains from the domestic pig (Webster, Hinshaw & Bean, 1977). Adaptation of such viruses to survival in the human species and the rapid achievement of global prevalence in the human population has not been recorded.

Natural reassortment of genes from different influenza A viruses

Novel strains infecting man might also arise by reassortment of genes from human strains with genes from influenza A viruses infecting non-human hosts. Reassortment requires that a single host individual shall be infected simultaneously with virions of two or more different major serotypes. Reassortment of influenza A virus genes occurs readily in nature, especially in birds, in which several strains of influenza A virus commonly co-infect single birds (Webster & Campbell, 1974); Scholtissek, Koennicke & Rott, 1978; Desselberger *et al.* 1978; Gardner & Shortridge, 1979; Hinshaw *et al.* 1980).

Reassortment can be produced experimentally in non-human hosts and the hybrids remain genetically stable on transmission (Webster & Laver, 1975; Arikawa, Yamane & Ishida, 1981). Reassortment occurs naturally in humans simultaneously infected with two strains of influenza A virus (Yamane *et al*; Young & Palese, 1979, Nishikawa & Sugiyama, 1983).

Reassortment and recombination with strains from non-human hosts is therefore available under natural conditions for the construction of novel major serotypes of human influenza A virus.

A pandemic strain has been made to survive in an alternative host species by a recombination that changed the host range, and it was then made to regain the original host by a second recombination (Scholtissek, Koennecke & Rott, 1978). The experiments involved the virus of fowl plague growing in cell culture, and demonstrate a possible mechanism for the origin and maintenance of recurrent major serotypes in man, but it is difficult to envisage how the findings can explain what has actually been happening in human epidemic influenza.

Related strains in human and non-human hosts

If humans are deriving major influenza A serotypes from viruses parasitizing other species of host, evidence should be found in similarities in the viruses from the two sorts of host. Recombinants should reveal similarities in the gene coding for the haemagglutinin and perhaps also in that coding for the neuraminidase, because these genes account for antigenic shift.

In 1968, when human H2N2 strains were superseded by H3N2 strains, only the gene coding for the haemagglutinin had been changed by a reassortment to form the A/Hong Kong/1/68 virus, and the new haemagglutinin was closely related to that of A/duck/Ukraine/1/63 and to that of A/equine/Miami/1/63 (Ward *et al.* 1981). The 1968 human H3N2 virus and the 1963 viruses infecting ducks and horses all possess a haemagglutinin that may have been derived at some time from a common ancestor virus. The haemagglutinin of the 1963 duck virus differs in 23 amino acid sequences from that in the human virus of 1968 (Ward & Dopheide, 1981; Fang *et al.* 1981).

Human influenza A viruses of the H0N1 and H1N1 (old style) eras, 1930–56, possess a neuraminidase similar to that in Hsw1N1 strains from swine and to that in the avian strain, A/duck/Germany/1868/68.

In 1957 the novel human haemagglutinin resembled that of an avian virus, A/turkey/Massachusetts/65 (Webster & Laver in Kilbourne, 1975, pp. 306–7).

Instances of such relatedness will probably continue to be discovered, and they are evidence of aspects of the evolutionary history of influenza A viruses as

https://doi.org/10.1017/S0950268800066851 Published online by Cambridge University Press

parasites of man and other hosts, but they do not necessarily indicate a process whereby antigenic shifts are currently occurring in the viruses affecting the human host species.

Observations favouring anthroponotic derivation of human major serotypes

Recycling of major serotype eras

A single major serotype of human influenza A virus (H0N1) caused all the type A influenza cases verified by virus isolation from its discovery in 1933 until 1946. Serological studies suggest that its era of world dominance had begun about 1929, at the end of an era of human Hsw1N1 strains that began in 1918. H0N1 strains were replaced worldwide by H1N1 (old style) strains from 1946 until 1957 when they in turn were replaced by H2N2 strains. In 1968 H2N2 were replaced by H3N2 strains, which caused almost all recorded influenza A cases until 1977. Thus there were four and possibly five eras of worldwide dominance by viruses belonging to single major serotypes. In 1977 H3N2 strains were joined by a recurrence of an era of H1N1 (old style) strains, and the two are still co-circulating in 1987.

Sera collected before the appearance of H2N2 virus in 1957 showed that many persons born before 1908 already possessed antibody against the novel virus antigens. Similarly in 1968, pre-epidemic sera from some older persons were found to contain antibody against H3N2 virus. Some 90% of persons born between 1857 and 1877 possessed measurable H2N2 antibody (Mulder & Masurel, 1958; Schild & Stuart-Harris, 1965; Masurel & Marine, 1973; Ivannikov *et al.* 1980; Rekart *et al.* 1982). The findings supported an earlier suggestion that human influenza A virus might possess only a limited number of effective antigens, and that these might be being successively recycled as a strategy for survival of the influenzal parasite in the human host species. These sero-epidemiological studies indicated that a sequence of earlier major serotype eras had occurred that was comparable with those that have continued since the advent of virology (Davenport, 1977). H2N2 viruses seem to have been dominant during the last quarter of the nineteenth century and to have been succeeded by H3N2 strains from 1900–17. The succession of Hsw1N1 and H0N1 has already been described (Langmuir & Schoenbaum, 1976).

These tentative suppositions about a pattern of recycling received corroboration when, in 1977, H1N1 (old style) viruses reappeared after 20 years' absence and rapidly again established world distribution and an era of prevalence. Serological evidence suggests that H1N1 (old style) strains had previously circulated from 1908–17 during the earlier prevalence of H3N2 viruses (Masurel & Heijtink, 1983).

Detailed examination of H1N1 isolates of 1977–8 reinforced the anthroponotic concept, because they were homogeneous and were identical with the Scandinavian variant that had caused much of the influenza in the great epidemic of 1950–1 and had again been epidemic in 1953. They differed from the earliest (1946) and from the latest (1956) strains of the earlier H1N1 era (Kendal *et al.* 1978; Scholtissek, v. Hoyningen & Rott, 1978; Nakajima *et al.* 1979).

After an absence of 25 years this Scandinavian-like strain re-established itself within a single season as a prevalent serotype in many parts of the globe, at first confining its attack largely to persons too young to have encountered its previous

prevalence. The homologous protection resulting from exposure to H1N1 (old style) viruses more than 20 years earlier did not depend upon the presence of strain-specific haemagglutination-inhibiting antibody in the persons exposed to reinfection (Gill & Murphy, 1985). Immunity may be life-long against reinfection by a virus of the same serotype, though the degree of protection may vary with the major serotype. Virtually all non-immune persons in the world population except those in almost inaccessible communities are infected during each era of major serotype prevalence.

Where and in what condition was the Scandinavian-like strain of H1N1 (old style) influenza virus persisting for a quarter of a century? What reactivated it? How did it achieve worldwide dissemination in the human population within a few months? It must have been dormant either in the human or in some non-human host species. Absence of antigenic change in HA or NA seems to exclude continuous low-grade transmissions in man, yet the absence of H1N1 (old style) virions in other host species favours the hypothesis of human endemicity, as does the finding that the 1977 H1N1 strain is identical with A/FW/50 (H1N1) that caused worldwide epidemics from 1950 to 1953.

Some experienced observers have suggested that the reappearance of A/FW/50 (H1N1) strains in 1977 does not represent a stage in the natural behaviour of influenza A viurs, but a human error allowing release of a laboratory-held strain. If that were so, one would be faced with the seemingly insuperable difficulty of explaining how such a laboratory escape could achieve worldwide distribution within a single season.

The antigenic drift of H1N1 strains since 1978 differs entirely from that occurring during the earlier prevalence after 1953 (Yakhno *et al.* 1981). Two findings deserve especial mention. First, sequential mutations in the later H1N1 strains after 1977 were scattered throughout the genome as commonly as in the genes for the surface proteins HA and NA, therefore selective antibody pressure cannot be solely responsible for the emergence of minor antigenic variants. Secondly, although the 1977 H1N1 viruses arose from the 1950 strain, subsequent H1N1 isolates in the 1978–79 season arose by recombination with co-circulating H3N2 strains. The genes for HA, NA, M and NS came from the H1N1 parent, so maintaining the H1N1 serotype. Evidently genetic variation of strains within a major serotype is not restricted to mutation but can occur by genetic reassortment (Young, Desselberger & Palese, 1979), so it is perhaps unwise to base hierarchical classification of influenza A viruses solely on criteria of mutation or reassortment.

Anachronistic findings

The discovery of particular strains of influenza A virus during times of their supposed absence is of great theoretical significance. Anachronisms may take several forms. The commonest is when prevalent strains are found in the absence of an epidemic, as in 1954 when Zakstel'skaya isolated H1N1 (old style) strains from healthy persons when no influenza was being reported (in Zhdanov, Solov'yev & Epshsteyn, 1960).

Minor variants are sometimes isolated from influenza long after they had disappeared, yet still within the prevalence of their major serotype. For example in 1979 in Adelaide, Australia, a 3-year-old boy's attack of croup was caused by

A/Aichi/2/68, an H3N2 strain last isolated 10 years previously (Moore *et al.* 1981, and see next section).

Anachronisms have been found during the long periods that separate the recurrent eras of prevalence of the same major serotype. Thus H1N1 strains were isolated in 1962 during the 1957–68 era of H2N2 dominance. Although they differed from both the earlier (1946–56) and the later (1977–) strains, they were conserving the H1N1 (old style) major serotype in the human population. They contained no gene of the then current H2N2 virus (Klimov & Ghendon, 1981). Again in 1980, 12 years after the end of the 1957–68 era of H2N2 prevalence, H2N2 strains were isolated that had haemagglutinins and neuraminidases similar to those of A/Singapore/1/57, absent for 23 years. Three of these 1980 H2N2 strains compared with the 1957 H2N2 reference strain showed no polypeptide difference by electrophoretic mobility in PAGE, or mobility of duplexes by hybridization of the virion and cRNA of A/Leningrad/553/80 and A/Singapore/1/57. The haemagglutinin of A/Leningrad/549/80 resembled that of 1957 strains but not that of 1954 strains. They were, however, distinguishable from H2N2 strains of the 1957–68 era by HI tests using sera from infected polecats, the 1980 H2N2 strains being 'older' antigenic mutants than the 1957 strains. They differed from laboratory strains and are therefore genuine H2N2 anachronisms (Golubev *et al.* 1984; Golubev *et al.* 1985).

In 1982, during the joint eras of H3N2 and H1N1 (old style) viruses, 0·9% of 652 sera from Leningrad children were found to contain antibody against H2N2 virus, and 1·2% contained antibody against H0N1 virus. Paired sera from 247 of them showed seroconversion to the haemagglutinin and neuraminidase of an H2N2 strain in 2·0% , and to those of an H0N1 strain in 0·4%. None of these children had been born until after the H0N1 and H2N2 eras of prevalence so, although the numbers are small, the findings show that viruses of these past major serotypes continued to be present in the Leningrad population (Ivanova *et al.* 1982).

Yet another sort of anachronism must be mentioned, an anticipatory change in antigenicity of the haemagglutinin. For example, the haemagglutinin of Dutch/1956, a late H1N1 (old style) variant, was more closely related to that of early H2N2 strains than to that of earlier H1N1 (old style) strains (Gengqi *et al.* 1980). Similarly, during the last years of H2N2 prevalence in 1966–8, antibody to H3 antigen was found to be gradually increasing in the population and Hsw1 titres were also found in the sera of children. In 1976, in pre-vaccination sera, Hsw1N1 antibody content was even greater than in the 1966–8 sera.

Sera from unvaccinated children, taken in 1976–7 before H1N1 (old style) virus had reappeared in 1977, contained with increasing frequency antibody to H1N1 (old style) virus (Monto & Maassab, 1981). These findings were interpreted as indicating that antigenic shift must be arising within the human population, or perhaps in some cases from swine..

Influenza in remote communities

Communities with few contacts with the outside world have provided valuable, sometimes puzzling information about the epidemiology of influenza. Anachronistic viruses have sometimes caused epidemic influenza in remote communities,

as in 1949 when the population of Alaska was attacked by H0N1 strains whose era of world prevalence had ended 3 years before (Fenner & White, 1970).

In 1972 some Amazonian tribes had had so little contact with the outside world that the sera of many adult Indians possessed no antibody against any known influenza virus (Black *et al.* 1970). However, in June 1972, among the 192 Indians composing the Mekranoti tribe, 59/61 sera were positive with H2N2 antigen and all were negative with H0N1 antigen. Only a few of the Mekranoti had titres, albeit low ones, to H3N2 stains which were already in their fourth year of world dominance. Those Indians with H2N2-positive sera were of all ages including children under 2 years old. Thus H2N2 viruses must have caused an epidemic in the Mekranoti community during the 1970–1 season, 2 years after being displaced from world dominance by H3N2 viruses (Napiorkowski & Black, 1974).

Records dating from before the discovery of human influenza virus in 1933 must be accepted with caution, but they sometimes describe influenza epidemics so excellently that they should not be altogether discounted. Shipboard outbreaks in the days of sail are of particular interest because the crews were often at sea for weeks or months. The fleet of Admiral Kempenfeldt sailed from Spithead on 2 May 1782, and remained at sea until influenza broke out with such intensity at the end of May that the ships were compelled to return to port early in June. Hirsch (1883) gives details of this and similar nautical outbreaks, and describes epidemics of influenza in remote communities after the rare arrival of a ship. 'The fact itself can hardly be doubted; while the striking thing appears to me to be that the strangers themselves, in all the cases, have remained exempt, or almost exempt, from the epidemic.' Such a phenomenon would be expected if the disease were being spread by symptomless carriers. A virologically confirmed example occurred at Point Barrow, Alaska, in 1935 when three healthy travellers arrived by plane from Canada, and 8 days later an epidemic of influenza began among the inhabitants (Burnet, 1945).

Seeding of influenza virus and herald epidemic waves

It has often been suggested that the behaviour of epidemic influenza cannot be explained without invoking some process whereby the virus is widely seeded throughout the world population before the epidemic (e.g. Langmuir & Schoenbaum, 1976). Evidence of such a seeding process was obtained in Atlanta, Georgia, USA, when in the non-epidemic season of 1972–3 H3N2 strains were isolated that were antigenically identical with those that caused the epidemic in the subsequent season. The authors suggest that the epidemic was produced by the potentially epidemic strains that they had found in the population during the previous season, and not by re-introduction of the virus in the 1973–4 season (Marine, McGowan & Thomas, 1976).

The prophetic serology mentioned towards the end of the previous subsection may be connected with the phenomenon of seeding. Monto & Maassab's findings (1981) and those of Gengqi *et al.* (1980) showed that by a rise of antibody a future dominant HA may cast its shadow before it. In 1975 the H3 antigenic determinant was becoming poorly manifest in the H3N2 strains as compared with those isolated in the early years of the era beginning in 1968 (Rovnova *et al.* 1978).

Shope (1958), finding pre-epidemic antibody only in persons over 70 years old,

survivors of an earlier H2N2 pandemic, speculated that the virus may be persisting masked in the human respiratory tract and may thus be widely seeded in the world population. Wallace (1977) made a similar speculation on analogy with the epizootiology of influenza A virus in swine.

Continuous monitoring is needed to reveal that a small wave of influenza viruses isolated in the latter half of one epidemic may herald next season's virus. During the herald wave only 0·4–2·0 % of respiratory infections yield the prophetic strain (Glezen, Couch & Six, 1982), proportions similar to those of the anachronistic findings mentioned earlier.

Obstacles to interspecies transmission and natural genetic reassortment as causes of recurrent cycles of major serotype eras in mankind

Numerous difficulties attend the successful transmission of influenza A virus from other hosts to man, and its subsequent adaptation to survival as a parasite of mankind. The difficulties may be extrinsic, e.g. geographical or temporal segregation of hosts and habitats, or intrinsic, e.g. mutual adaptation in the host–parasite relationship, especially at the cellular level.

Among the influenza A viruses co-circulating in birds within a region, genetic reassortment is restricted to highly related genes. Moreover, reassortment with intrusive genes of the influenza A viruses in birds from other regions is so rare that free distribution of genes by migrating birds may not occur easily. Intrusive polymerase genes may be unable to compete with the local alleles. Body temperature of the host may also be a selective character (v. Hoyningen-Huehne & Scholtissek, 1983). Nevertheless, avian influenza viruses appear to cross species barriers more readily than the influenza viruses parasitic in other host species, and wild birds have been proposed as the primary source of all influenza A viruses, mankind being seen as one of the sources of secondary spread (Alexander, 1982).

The host range of the virus seems to be determined by a single gene, Gene 1, that codes for the polymerase-associated protein in P3 (Almond, 1977). The transposition to life in a different cellular environment is not a simple affair, because the adaptive changes are not confined to the virus, the host cell too being progressively altered in a co-evolution between virus and cell (Ahmed *et al.* 1981). Indeed, the host cell is itself one of the determinants of the mode of parasitism (Conti *et al.* 1980; Patterson & Oxford, 1986). Antigenic changes in the influenza virus haemagglutinin may cause changes in the receptor binding site for host cells that have a deleterious effect on virus binding, and thus on infectivity and reproduction. Such mutual deleterious changes may explain the progressive decline in severity of successive epidemics in the era of a major serotype and the limited timespan of the era. They do not, however, explain the mechanism whereby the major serotype is sequestrated for many years and then recycled (Whittaker & Underwood, 1980).

Reassortants of human and avian influenza A virus containing the human virus gene coding for the haemagglutinin cannot replicate in the gut of ducks without developing two mutations in the receptor binding site of the haemagglutinin (Naeve, Webster & Hinshaw, 1983, 1984). The haemagglutinin receptor is highly species-specific (Rogers & Paulson, 1983), as might be anticipated on evolutionary grounds (see also Rogers *et al.* 1985).

If antigenic shift resulted from the recombination of influenza A viruses from a human and a non-human host it would be expected that highly virulent strains could be prepared by recombining viruses isolated late in the pandemic era with viruses obtained from mammals or birds. This is not the case. The high epidemicity that human influenza A viruses have acquired during their direct selective adaptation to man plays a leading role in their spread throughout the world population. Novel antigenic structure plays a secondary role by increasing the proportion of non-immune subjects in the population.

RNA–RNA hybridization techniques indicate the existence of five ribonucleoprotein groupings of influenza A viruses in different host species, namely (*a*) most avian strains, (*b*) some strains from seagulls, (*c*) recent strains from horses, (*d*) only A/equine/Prague/1/56 and (*e*) all the human and swine strains. 'These proteins have evolved functionally significant differences that favour their replication in a specific host' (Bean, 1984). Different techniques may thus reveal relationships between major serotypes. A close relationship between human and influenza A viruses belonging to the Spanish (1918–29) and the Singapore–Hong Kong (1957–) eras was most clearly demonstrated by testing the intranasal cross-protection of immunized mice (Smorodintsev *et al.* 1981*a*).

The prevalence of the virus in ducks is geographically variable. In Alberta many more ducks harboured virus than those in migrant flocks in Tennessee, and the prevalence in domestic ducks from an area of the People's Republic of China was fivefold that of those raised in Hong Kong (Gardner & Shortridge, 1979).

ASPECTS OF THE BEHAVIOUR OF HUMAN INFLUENZA A AND ITS VIRUS FOR WHICH THE NEW CONCEPT MAY PROVIDE THE EXPLANATION WHEREAS THE MEASLES MODEL CANNOT

Problem list

Ubiquity. Epidemics of influenza A occur in human communities worldwide, often in a single epidemic season – a mode of behaviour that excludes the primary epidemiological importance of climate, weather, temperature, humidity, geographical location, altitude, ethnic factors and social conditions as well as direct spread from the sick.

Seasonal epidemicity. Direct spread cannot, without additional suppositions, explain the seasonal occurrence of epidemic influenza A.

Antigenic drift of influenza A virus.

Disappearance of the prevalent strain.

Prompt replacement over the area of prevalence by the drifted variant(s).

Inter-epidemic survival of influenza A virus during apparent absence.

Explosive epidemics involving vast populations.

Timing of epidemic phenomena and *identity of strain* in small localities contempraneously with those in the whole country.

Cessation of epidemics in situations favourable for continuance by direct spread from the sick.

Absence of demonstrable serial interval dividing causal (introducing) influenza A cases from the cases that they have caused.

Low secondary attack rates in household outbreaks.

Anomalous age distribution of persons attacked.

Antigenic shift of major serotype.

Disappearance of prevalent major serotype.

Prompt worldwide replacement by successor major serotype.

Recycling of major serotypes after many years' absence.

Virological and serological anachronisms

Seasonal timing of antigenic shifts and antigenic drifts.

Annual flow of epidemic influenza A north, south and north within the world population across the surface of the earth.

No apparent secular change in behaviour of epidemic influenza throughout the last four centuries despite enormous increase in population, human contacts, and speed and complexity of human communications.

Doubtless there are problems that have been omitted. Nevertheless it is a formidable list calling for a reasonable and unifying explanation. The following subsections describe how the new concept of epidemic process seems to provide a relatively simple and satisfying explanation of almost all the difficulties that are inexplicable by the current direct-spread hypothesis. Where two or more of the problems appear to be linked they are considered together in the same subsection.

Influenza A is both seasonal and ubiquitous

Epidemic influenza attacks human communities all over the world except for some that are too remote and isolated, and novel variants of influenza A virus appear in many parts of the globe within months of being first discovered.

Epidemic influenza is seasonal, outbreaks in particular localities occurring almost annually at approximately the same season (Hope-Simpson, 1981). The epidemics occur in the colder months both north and south of the equator. Familiarity may have blinded us to the crucial epidemiological significance of the seasonal nature of influenza. A winter factor has been invoked to explain the epidemic mechanism (Andrewes, 1958), but climate cannot provide the explanation because influenza attacks communities living in diverse climates from the tropics to subpolar regions.

The global picture contrasts with the episodic local picture. Influenza is continuously travelling smoothly across the surface of the earth north, south and north again every 12 months, causing epidemics south of the tropics about 6 months before and after those north of the tropics, and occurring within the tropical belt twice yearly around the time of equinoxes. This annual swing of epidemic influenza to and fro across the whole earth was noticed long ago and was compared with the behaviour of certain migratory birds. Arctic terns make a similar annual grand journey (Andrewes, 1952) and it was later found that terns do in fact harbour influenza A viruses.

Influenza is among numerous seasonal phenomena whereof the seasonal mechanisms have not yet been elucidated, but certain principles which apply to all seasonal phenomena are relevant to influenza. The seasons are caused by a difference of about $23\frac{1}{2}°$ between the plane of daily rotation of the earth and the plane of its annual orbit around the sun. Were the planes the same there would be no seasons because vertical solar radiation would fall monotonously upon the equator. The tilted plane of earth's daily rotation causes vertical solar radiation to take a sinuous annual journey through the tropics, reaching the tropic of Cancer in the north on 21 June and the tropic of Capricorn in the south on 21 December and

crossing the equator twice yearly at the equinoxes. One is struck by the similarity to the annual path pursued by epidemic influenza albeit 6 months later. The great variations in solar radiation falling on different parts of the earth's surface at different times of the year give rise to all the diverse seasonal phenomena in living and non-living things. There is thus an inescapable natural law that, whatever the mediating mechanisms: *all seasonal phenomena ultimately result from these variations in solar radiation caused by the tilt of the earth.* Epidemic influenza cannot escape this law, so that no concept of the epidemiology of influenza can be accepted as valid that does not include the influence of season. Solar radiation has many components which, singly or combined, produce effects upon the earth, and the potency of their variation is everywhere apparent. Seasonal changes in many animals and plants are mediated through variations in temperature, humidity and photoperiod. The mediating mechanisms controlling seasonal epidemicity of influenza have not yet been elucidated, indeed they have hardly yet been sought (Gneyshev & Oi', 1977), but they exist and must be ultimately traceable to this inexorable extraterrestrial influence.

Epidemicity is not the only seasonal feature of type A influenza. Antigenic changes in the virus, both major and minor, occur seasonally and must also be included in any epidemiological hypothesis. Indeed, two such fundamental characteristics, epidemicity and antigenic variation, must be suspected of being two aspects of a single seasonally determined epidemic process affecting the virus.

The influence of season is integrated into the new concept by proposing that a seasonally mediated stimulus reactivates the virus residues latent or persistent in human host carriers. The seasonal character of influenza A epidemics worldwide is thus simply explained, because epidemics would have the opportunity to develop around such carriers in the wake of the seasonal stimulus that had recalled their symptomless persistent infection to infectiousness. The seasonal timing of antigenic drift is also explained, as discussed later.

Our understanding of the behaviour of epidemic influenza A is much altered if the new concept be correct. The travelling of influenza A south and north across the globe each year, inexplicable by direct spread, is not caused by movement of the virus from person to person, but results from the inexorable annual swing of the seasonally mediated stimulus provoking ubiquitous carriers to become infectious and able to infect their non-immune companions. We are witnessing a seasonal crop, some seasons favouring, others being less favourable for the crop of influenza cases (Hope-Simpson, 1981).

Survival of influenza A virus between epidemics

In 1944 Burnet (1945) concluded that there is 'no visible alternative to the view that human influenza viruses survive between epidemic periods in the tissues of human carriers'. Thirty years later Kilbourne suggested that silent (inapparent) influenzal infections must be providing a continuous chain of transmissions whereby the virus survives from one epidemic to the next perhaps 2 or 3 years later (Kilbourne, 1975). There is, however, no evidence to support the hypothesis. Subjects of subclinical influenzal infection are not known to be capable of transmitting the virus.

The new concept proposes that the virus is lying persistent between epidemics

in the tissues of humans who have suffered an attack of influenza, and is reactivated to infectiousness by a seasonal stimulus. The epidemics develop among the non-immune persons in contact with these carriers.

The new concept rests on this proposition that the virus must be surviving between epidemics and eras of prevalence in some mode of inapparent human parasitism. Search is therefore needed by modern methods for persistent influenza virus and for latent residues of the influenza A virus genome in tissues of humans known to have been infected. One suspects that elderly persons would be the most promising in the search for latent residues, whereas younger persons might be better candidates for persistent infection. Negative and positive findings should both be published.

Antigenic drift, strain disappearance and rapid replacement

During successive epidemic seasons of the era of prevalence of each major serotype of influenza A virus, minor mutations occur in all eight genes. The mutations in the genes coding for the surface proteins haemagglutinin and neuraminidase give rise to minor antigenic variants of the parent strain, a process of antigenic drift. A strain that has been causing all the influenza A over a large area of the Earth will often disappear, and be replaced next season by a novel minor variant of the same major serotype. For example, A/Port Chalmers/1/73 (H3N2) strains, which had been causing all the influenza A in many parts of the world in the 1974–5 season disappeared, and A/Victoria/3/75 (H3N2) strains caused all the influenza in the same areas in the 1975–6 season.

Four problems call for explanation, as follows. What is the mechanism of antigenic drift? What causes the prevalent virus to disappear abruptly? How does the successor strain colonize vast, widespread populations within a few months? How can such a change take place between epidemic seasons when the virus appears to be absent?

Antigenic drift, caused by the interplay between viral mutability and human host immunity, is usually attributed to herd immunity, the selective pressure of antibody in a community partially immunized by the previously prevalent strains (Webster & Laver, 1975). There are two weaknesses in this superficially attractive explanation. First, if the sick person were transmitting the virus, the companions he is destined to infect would not be those already possessing immunity from infection by a strain of the prevalent major serotype, because such second infections are relatively infrequent. His victims would consist almost entirely of persons not immune to the current major serotype, persons therefore who could not be exerting any immune selective pressure. Secondly, he could not himself exert such immune pressure because during his illness, when he was presumed to be transmitting the virus, he would not have had time to develop his own specific immunity. These may be the reasons why measles virus in contrast to influenza A virus has remained so antigenically stable. When measles virus does meet the immunity it has provoked, as in persistent measles virus infections, variants of the parent virus are, in fact, produced.

The explanations offered for abrupt disappearance of widely prevalent influenza A virus strains and their rapid replacement by a minor variant are equally vague and unsatisfactory (Webster & Laver, 1975).

The new concept proposes that latent residues of the previously prevalent strain are later seasonally reactivated and so must inevitably encounter the immunity that the virus has itself engendered in the person in whom it had caused influenza. Reconstituted virions identical with the parent strain are therefore at a disadvantage within the host carrier in competition with the mutant particles that are always produced during an influenzal infection. During persistent infection, variants are produced even more rapidly. Particles identical with the parent virus are seen as being arrested within the carrier by his specific immunity to them, so that he can only transmit an assortment of mutant particles from which companions who are non-immune to the current major serotype unconsciously select the one(s) most fit (in an evolutionary sense) to survive and continue the major serotype. Recent findings show that the host cells of the carrier himself may be exerting even greater selection than was envisaged by the new concept as originally proposed (Patterson & Oxford, 1986).

It has been argued earlier in this paper that persistent infection is the mode of parasitism during which antigenic variation is most apt to occur, and the new concept accords with the laboratory methods used for selecting minor variants since the seminal observations of Archetti & Horsfall (1950) and Isaacs (1951). They showed that the Liverpool strain of H1N1 (old style) influenza A virus grown in chicken embryo in the presence of Liverpool antibody produces a harvest of the Scandinavian variant, and vice versa. The new concept proposes that influenza A virus reactivated after persistent infection in persons recovered from acute influenza A is subject to similar immune pressures that favour variants in competition with the parent virus. The concept thus explains not only antigenic drift but also the abrupt disappearance of the predecessor strain and its universal replacement next season by the successor by means of a stepwise change during the inter-epidemic periods (Hope-Simpson, 1979). All persons infected by the same strain would have developed a similar immunity and would have offered their companions a similar assortment of mutants, of which one in particular would usually have been the successful competitor. Thus in the 1975–6 season A/Victoria/3/75 (H3N2) seems to have been the fittest mutant from the A/Port Chalmers/1/73 (H3N2) parent strain of the previous season, and it accordingly replaced the Port Chalmers strain throughout the large proportion of the world population in which it had caused influenza. The suggested mechanism explains the apparent metamorphosis that had taken place during the inter-epidemic absence of influenza.

In some seasons more than one mutant of good evolutionary potential is produced and two or more may co-circulate. From two small brothers with influenza sharing the same bed in 1968, one yielded A/England/68 (H2N2) and the other A/Tokyo/67 (H2N2) strain (Hope-Simpson, 1979). Circumstances in different localities sometimes favour one variant as against another, as in the great 1950–1 epidemic, when Scandinavian and Liverpool variants of H1N1 virus co-circulated in different parts of the world.

Explosive epidemics

Influenza epidemics differ in character. Little protracted epidemics may last for several months even in small communities in contrast to vast explosions of the

disease which may attack 15% or more of a large community within 6 weeks and then cease. The first H3N2 influenza A virus epidemic in Great Britain in the 1968–9 season exemplifies the first sort, and the second in 1969–70 season provides the contrast. Explosive epidemics have long been seen as a challenge to the measles model of direct spread of influenza from the sick (Glass, in Thompson, 1852; Hirsch, 1883). Huge populations living in wide geographical areas have been attacked almost simultaneously after many months during which no influenza has been apparent, and no contact could be traced between the earliest cases. It will not suffice to say that the virus had previously been sustained by chains of symptomless infections without providing evidence that this has in fact been happening, and without providing suppositions explaining the further mechanisms that must exist to bring about the explosive changes alternating with periods of subclinical infections.

The new concept regards each seasonal outbreak of influenza A as a crop, the seeds of which are sown by symptomless human hosts who are always everywhere carrying latent influenza A virus. Some seasons favour, whereas others are less propitious to a good crop of influenza cases. In each season the harvest depends in part on the operation of the seasonal reactivating influence upon the carriers, and in part on their distribution and on that of their non-immune companions at the time of local reactivations of the virus (Hope-Simpson, 1981). Presumably the potentialities of the successful virus variant also affect the features of the epidemic.

Coincident timing and virus identity in small locations and in the whole country

Successive outbreaks of type A influenza in small relatively remote communities often coincide closely season after season with those of the country as a whole and, although the virus changes, the identical strains of virus appear contemporaneously in the two situations (Hope-Simpson, 1979). Such close concordance of the findings in small localities with those in the whole country could not occur if each successive variant of the virus, major or minor, had to invade the country from outside and attack the population afresh by direct transmissions from the sick. Many observers faced with such findings have admitted that independent foci of infection must be pre-existing in different countries. Such an explanation agrees with the new concept but underrates the number of such foci, and so it is inadequate to explain the findings and also requires further suppositions about the details and mechanisms of such pre-seeding and of the activation of the seeded virus.

On the new concept the world population is already seeded with carriers and the timing and pattern of both local and national outbreaks depend on the timing of the seasonally mediated reactivating stimulus and the distribution of carriers and their non-immune companions, who are providing the conditions for outbreaks to occur. The contemporaneous appearance of novel viruses in small and large areas is simply explained by the mechanism of antigenic drift proposed earlier. The finding that new major serotypes also appear contemporaneously in small localities and in the country at large indicates the operation of a similar process in antigenic shift and will be discussed later.

Cessation of epidemics despite abundant available non-immune subjects

The novel H2N2 influenza A virus caused a large epidemic in Great Britain in late September and early October 1957, and despite the presence of abundant non-immune persons, it then ceased, after having attacked some 15% of the immunologically inexperienced population. The next major serotype, H3N2, caused illness in only about 5% of those communities when it first appeared in England in January 1969. Why should such a highly infectious virus terminate its epidemic spread in situations so favourable for continued direct transmission from the sick? Measles does not similarly spare an immunologically virgin community. The explanation given – namely that the 5% who were attacked had been only the visible tip of an iceberg in a population of which some 40% had been subclinically attacked – must have been incorrect because a much larger epidemic followed only 8 months later (Hope-Simpson, 1979).

Andrewes (1951, 1958) has proposed that overt illness and solid immunity follow the receipt of a large infecting dose of the virus, whereas persons receiving a small dose do not develop influenza and have only a transient immunity. His hypothesis has the added attraction that it also offers an explanation of antigenic drift and that it explains certain findings in the population antibody levels between epidemics. It fails, however, to explain the cessation of the first H3N2 epidemic, the survival of the virus between epidemics and the explosive onset of the second H3N2 epidemic.

Virus can be isolated from persons suffering from influenza and has occasionally been isolated from healthy persons, but it is proposed that virus is not naturally shed from the sick to infect companions during the illness.

By the new concept, on which each epidemic consists of the persons who have been infected from the symptomless carriers, the epidemics must cease automatically because the sick are unable to transmit the virus.

Absence of serial interval in cumulated household outbreaks

The only epidemiological evidence that an infective agent is being directly transmitted from the sick is the demonstration of the interval between the causal cases and the cases to which it has given rise – the serial or transmission interval or generation time. It can be demonstrated by cumulating household outbreaks using day 0 for the first day of the first case in each household. In a directly transmitted disease the falling curve of co-primary (introducing) cases is followed by a wave of secondary cases and sometimes by subsequent waves of tertiary cases, etc. (Hope-Simpson, 1948). Cumulation of households attacked in the first two epidemics caused by H3N2 viruses showed no serial interval (Hope-Simpson, 1979).

This is the result that would be expected if the new concept is correct. The introducer of the virus into each household would have been a symptomless carrier, not a case of the disease. All the cases were therefore secondary to an invisible primary. The absence of a serial interval, therefore, supports the new concept by providing evidence that direct transmissions are not (or not commonly) occurring from the sick.

Low secondary attack rates within households

Several observers have found low household secondary attack rates even in severe influenza epidemics, in contrast with high rates in such institutions as military barracks and residential schools (Davis *et al.* 1970; Hope-Simpson, 1979). An accurate household secondary attack rate can only be estimated by using the serial interval to differentiate secondary from primary and tertiary cases. In the absence of a known serial interval the 'subsequent attack rate' was employed by arbitrarily choosing a first-day case in each household as the introducing (primary) case and considering all others as secondary to it. The method would artificially overestimate the rate if the disease were being spread by direct transmissions, because all co-primaries and tertiaries would be counted as secondaries. Nevertheless, in the first H3N2 epidemic the subsequent attack rate in households was found to be only 17% and in the much larger second H3N2 epidemic only 14% (Hope-Simpson, 1979), ridiculously small values when compared with measles (75%), varicella (61%) and mumps (35%) (Hope-Simpson, 1952).

If, however, as proposed by the new concept, each introducer was not a sufferer from influenza but a symptomless carrier of reactivated virus, the rates in the first two H3N2 epidemics must have been 25 and 55%, not 17 and 14%. The revised values accord well with those reported from institutional outbreaks, in which no account can be taken of the introducing cases.

Anomalous age distribution of persons attacked in influenza A epidemics

An immunizing disease like measles in an immunologically virgin community attacks persons of all ages indiscriminately, but thereafter the average age of those attacked in subsequent epidemics is lower than that of the whole community because the infectious agent attacks chiefly younger persons born since the previous epidemic (e.g. Panum, 1940). Influenza caused by each major serotype usually confers life-long immunity against viruses of the same major serotype although the immunity is less solid than that conferred by measles. During each era of prevalence of a major serotype of influenza A virus those attacked in the later epidemics would be expected to be on average younger than the victims of the first epidemic. Influenza A virus has been found not to behave in this way. For example, H2N2 strains, which at their first appearance in 1957 had attacked almost indiscriminately persons of all ages, were found to be maintaining a similar broad age distribution in the last four of their eight epidemics in the same small population. The average age of those attacked in the final epidemic (1967–8) was almost the same as that of the whole community and ranged from 10 months to 86 years (Hope-Simpson, 1984). Although pre-school children suffered the highest incidence rate, neither they nor school-aged children appeared to have been specially important in transmitting the disease. Successive epidemics caused by a directly transmitted immunizing infective agent should not have produced repeatedly this almost indiscriminate age pattern in a small community.

The picture is, however, comprehensible on the new concept. Each epidemic would consist only of the non-immune companions in effective contact with carriers in whom latent virus was being reactivated, so that each epidemic would have been automatically self-limiting, even the first one in a non-immune popu-

lation. The whole era of prevalence of a major serotype would thus resemble a single measles epidemic, each successive influenza epidemic corresponding to only one generation of the measles epidemic.

A simple model of the new concept shows that the persons infected from carriers tend to comprise a vertical slice through the age structure of the community in each successive epidemic (Tables 5–8 in Hope-Simpson, 1984).

During their era of prevalence, viruses of a major serotype probably attack almost every member of the world population except those persons old enough to have suffered during an earlier era of the same major serotype, and those too remote from human communications such as Amazonian Indian tribes (Napiorkowski & Black, 1974) and remote island communities (Mantle & Tyrrell, 1973). The new concept suggests how influenza A viruses may have evolved a host–parasite adaptation in man whereby it is enabled to parasitize almost the whole world population without destroying its own basis of survival.

The return of H1N1 (old style) strains in 1977 after only a score of years' absence provided an illustration of how a whole era of prevalence of a major serotype of influenza A virus resembles a single measles epidemic. Throughout the world the persons attacked by H1N1 strains in 1977 and 1978 were almost entirely those born since the previous era of prevalence of H1N1 strains (1946–57), as would have been the case with measles returning after 20 years' absence. Recurrences of major serotypes after longer absences have been discussed earlier.

The behaviour of epidemic influenza A does not seem to have altered with the increasing speed and complexity of human communications

Hirsch (1883), when discussing the question of contagiousness of influenza, wrote:

> In more recent times the great majority of observers have answered it decidedly in the negative, not so much on the many single observations which tell against the communicability of the disease, as on the ground that the spread of influenza can be shown to have taken place quite independently of intercourse...It has not spread more quickly in our own times with their multiplied and perfected ways and means of communication, than in former decades or centuries.

The remarkable rapidity with which influenza has involved vast populations has elicited amazement throughout medical history. Hirsch gives examples and quotes Jones writing in Philadelphia in 1826, that

> this epidemic affects a whole region in the space of a week, nay, a whole continent as large as North America, together with all the West Indies, in the course of a few weeks, while the inhabitants could not within so short a time have had any communication or intercourse whatever across such a vast extent of country. This fact alone is sufficient to put all idea of its being propagated by contagion from one individual to another out of the question.

Hirsch's discussion of the problem is thought-provoking. Lines of communication are now more difficult to trace because the population is denser and more peripatetic and communications far more 'multiplied and perfected' and rapid than in 1880, so that current observations purporting to record epidemic movements are difficult to interpret.

In 1776 Fothergill circulated a questionnaire to a score of medical colleagues in

England, Wales and Scotland requesting details about the mild but widespread influenza epidemic of the last months of 1775. The replies (Thompson, 1852), by the timing of the epidemic in different areas, might well be describing a twentieth-century influenza epidemic.

Such morbidity records from previous centuries are scarce, and attention has therefore been directed to mortality data. Severe influenza epidemics sometimes impose a characteristic sharp elevation on the general mortality, which increases within 2 or 3 weeks to a peak well above the value expected for the time of year and then declines within a further 2 or 3 weeks to the expected value (Housworth & Langmuir, 1974; Alling, Blackwelder & Stuart-Harris, 1981).

If the disease were spreading directly from the sick it seems likely that the characteristic mortality peak signalling the recent visit of a severe influenza epidemic would not have occurred in previous centuries because of the retarding effect of poor communications in a sparser population. Analysis of parish burials in Gloucestershire, however, showed that a similar elevation of general mortality often occurred when an influenza epidemic in Great Britain was described in contemporary records (Hope-Simpson, 1983). Further studies confirmed the finding in other counties and showed that epidemic patterns of influenza in this country have not altered in four centuries (Hope-Simpson, 1986).

Such a finding is predicted by the new concept. The apparent speed with which the disease travels is not caused by the transmission of the virus but reflects the inexorable movement across the Earth of the seasonally mediated stimulus recalling to infectiousness the influenza A virus quietly persisting in ubiquitous carriers, and so affording opportunity for epidemics to spring up in its wake among their non-immune companions. The rapidity and complexity of human communications are irrelevant except in distributing the carriers and the non-immune subjects. In the present century the characteristic elevation of mortality follows hard on the heels of the severe influenza epidemic, and it appears always to have done so.

CONCLUSIONS

A valid concept of the epidemic process in human influenza A has much to comprehend in explaining the whole behaviour of the parasite in mankind. There are: the seasonal nature of epidemics, their origins and cessations, and the inter-epidemic absences of the virus; seasonal antigenic drifts and shifts arising from seasonal changes in viral molecular structure; the rapidity with which novel strains appear seasonally in vast areas or even worldwide. The currently accepted belief in measles-like spread directly from the sick is powerless to explain these familiar features and therefore provides no basis for a comprehensive hypothesis. The discovery of a genofond of influenza A virus in many non-human host species provided the promise of an alternative epidemiological concept, strengthened by the finding that genes originating from influenza A viruses from different host species could be recombined experimentally, and that such reassortment was occurring under natural conditions. This zoonotic explanation of antigenic shift received further credibility when it was found that the haemagglutinins and neuraminidases of certain human influenza A viruses were related to some of those

in the viruses of non-human hosts. The scientists who made these important discoveries were commendably cautious about claiming them as the explanation of the difficulties of influenza A epidemiology, but it now seems clear that influenza A viruses of man were related to those of non-human hosts during their evolutionary development, and that trans-species transmissions and recombinations still occasionally involve human hosts. Nevertheless the concept of direct spread of human influenza, even when fortified by the genetic reservoir of influenza A viruses in non-human hosts, is inadequate to explain the difficulties listed earlier.

On the other hand, the new concept advanced in this paper provides a relatively simple explanation of almost all the difficulties. It explains ubiquity, seasonal epidemicity, antigenic drift (reflecting laboratory experience), disappearance of the prevalent virus, prompt replacement by the successor(s), inter-epidemic survival of the virus, the differing natures (sometimes explosive) of epidemics, similar timing and viral identity in small and large areas, cessation of epidemics, absent serial interval, apparently low household attack rates, anomalous age distribution, seasonal antigenic changes in the virus, prompt distribution worldwide of novel strains, annual progression north–south–north over the globe, similarities of behaviour in modern and ancient times, and the presence of viral and serological anachronisms.

Hoyle, Wickramasinghe & Watkins (1986), in studying influenza epidemics in schools, considered that direct spread could not explain what was happening but concluded that influenza viral precursors must be reaching earth from an extraterrestrial source.

The new concept on the other hand sees the world population as almost everywhere always seeded with symptomless human carriers between successive epidemics during the era of the current major serotype(s), last season's sufferers carrying the virus as a persistent infection, the condition in which minor mutations most readily occur. During the long intervals between successive eras of prevalence it sees the major serotype maintained as latent subviral residues, a state in which changes in the structure of the virus are less likely to occur. During the epidemics the virus is seen as multiplying at a very high rate, rapidly producing defective interfering particles which lead it into the persistent infection state that ultimately leads on to latency. How the worldwide distribution of carriers originally came about is a matter of the evolutionary history of human influenza A virus, to which structural affinities with influenza A viruses of other host species doubtless indicate parts of the story. The annual trek of influenza A south and north across the globe is seen as the wake of the inexorable annual swing of the stimulus, mediated from seasonal variations in solar radiation, reactivating the virus in ubiquitous carriers, so allowing opportunity for epidemics to develop. A seasonal prevalence of any disease is the signal of some such extraterrestrial influence and it calls for a search for the mechanisms by which it is being mediated (Gneyshev & Oi', 1977). The ubiquity of human epidemic influenza A excludes as mediating mechanisms such local influences as climate, ethnic characters, social habits, altitude and humidity, though these may modify the presentation and course of the disease.

Does this new concept help to explain the phenomena attendant upon antigenic shift? Does it explain the abrupt disappearance of all strains of the major serotype that has been prevalent worldwide for more than a decade, or the manner in which

its successor achieves world distribution by next season, or the long period of world prevalence during which strains of that major serotype may be causing all the cases of influenza A, or the even longer absences between eras, or the recycling of strains of the major serotype after perhaps half a century of absence? Some of these features resemble those that characterize antigenic drift during a major serotype era. It is therefore not unreasonable to suppose that a concept that so simply explains the similar features of antigenic drift may also be applicable to the phenomena of antigenic shift. The necessary modes of parasitism of the virus exist.

The new concept depends upon the existence and ubiquity of symptomless carriers of persistent or latent influenza A virus.

> Very little can be said about the frequency of influenza virus carriers or the form in which the virus is carried. No systematic search for carriers has been reported, and it seems highly improbable that the present methods for the isolation of virus would be of any value for such a study. The mode in which the virus persists in the hypothetical carrier can only be a matter for speculation...Until some new approach to the problem develops, it seems unlikely that any evidence other than epidemiological will be available on the carrier question (Burnet, 1945).

Burnet's speculations remain unconfirmed, but much new information has accrued, and the evidence appears to favour the concept that major serotypes of influenza A virus are maintained between their eras of prevalence within the human host species (Smorodintsev *et al.* 1981*a*; Golubev, 1984). Man, it should be remembered, is the only domestic animal closely associated with mankind world-wide. If the new concept is correct, persistent infection is a normal mode in the natural relationship between influenza A virus and the human host. In cell culture the virus undergoes rapid antigenic changes during persistent infection until this phase is succeeded by that of latency of subviral residues which seem to be less mutable, and might remain lifelong in the human host, though liable to periodic reactivations as in cell culture. Whereas persistent infection may be involved in antigenic drift, latent subviral residues may maintain and recycle the major serotypes to cause antigenic shift. Findings in many parts of the world indicate that major serotypes are not altogether absent from mankind between their eras of prevalence, and their anachronistic epidemic appearance in remote populations indicates their potential to take advantage of a non-immune community (Golubev *et al.* 1985).

Whatever the primary origin of H3N2 virus it seems to have been pandemic around 1900–17, so there is no need to invoke the animal reservoir and recombination to explain its reappearance in 1968. Similar reasoning applies to the reappearance of H2N2 viruses in 1957 and of H1N1 (old style) viruses both in 1946 and in 1977 (Smorodintsev *et al.* 1981*b*). The order in which the major serotypes are recycled may be determined by the mechanism operating Francis' original antigenic sin.

The authors are aware of commonsense objections to the new concept, namely that virus obtained from the influenzal illness may be made to infect human volunteers, animals and laboratory cell systems, and that it seems to spread directly through the population. Subclinical infections seem to be occurring, as indicated by rises of antibody and occasional virus isolations from healthy persons.

The authors for many years attempted unsuccessfully to construct a valid

hypothesis of direct spread including such findings as indications of subclinical infections, and we are not aware that other workers have had more success. The field is still wide open.

It should, however, be remembered that if the new concept is correct healthy carriers will provide a rise in antibody in their sera when reactivation and renewed replication of virus is taking place, and one might be lucky enough to isolate the reactivated virus from such healthy persons.

The new concept, even if it is proved to be generally correct, does not exclude the possibility of direct spread from the sick also taking place, but not as the normal or main mode of influenza A virus transmission.

In order to authenticate the new concept virus needs to be found in human carriers between epidemics and subviral residues between eras of prevalence. Recent techniques with DNA–RNA and RNA–RNA probes may be helpful. The mechanisms mediating seasonal reactivation also need to be elucidated. Whether such investigations support or disprove the new concept, they will establish the epidemiology of influenza A on a more logical basis.

Meanwhile we suggest that there is a strong case for again re-classifying influenza A viruses to recognize each of the five major serotypes that have comprised the major variants of human influenza A (Smorodintsev, Golubev & Luzjanina, 1982). The 1980 classification, with continuous numeration H1–13 and N1–9 regardless of host species, conceals the main biological property of human influenza A viruses, namely their ability to produce global pandemics of influenza. It is as if one were to classify buildings by their building material rather than by their functions. On the other hand the 1979 WHO classification, identifying the five major HA serotypes in man – Hsw1, HO, H1, H2, H3 – provided an intelligible history of pandemic influenza A since 1918 and clues as to its earlier history. Epidemiology provides the evidence of the individual identities of Hswl, HO and H1 (old style) haemagglutinins in the human major serotypes, and the classification ought to match the realities of the behaviour of influenza A virus in man. Each of those three in succession imposed its own specific immunological memory on those it attacked, leaving them open to attack from one another rather than from kindred minor variants of the same major serotype. The Spanish era of H1N1 (new style), 1918–57, thus conceals three important eras corresponding to successive dominances of these three different influenza A viruses. The return in 1977 of H1N1 (old style) viruses supports the suggestion of Francis & Davenport and of Smorodintsev that the pandemic human influenza A viruses may perhaps be limited to these five major serotypes, and if so they will continue to be recycled.

We wish to thank Professor G. I. Karpukhin, Director of the All-Union Research Institute of Influenza, Leningrad, for help and encouragement in the preparation of this paper. Also Mrs Bettie Neal for secretarial help and Dr John Oxford for helpful discussions.

APPENDIX

The latency and seasonal-reactivation concept of epidemic process in influenza A

The following propositions are developed from those originally published in table 2 of Hope-Simpson (1979). They are offered tentatively for discussion and amendment.

Proposition 1, concerning latency of influenza A virus. During influenzal illness in the human host influenza A virus so rapidly becomes persistent in his tissues that he cannot normally transmit it to his companions. The persistent virus causes him no further illness. He develops specific immunity and becomes a life-long carrier host of the latent virus, at first as a persistent infection and later as subviral residues. He is not infectious to his companions except when his persistent virus or its residues are reactivated.

Proposition 2, concerning seasonal reactivation of latent influenza A virus. The persistent virus in carrier hosts is reactivated seasonally by a stimulus that, being ultimately dependent on the seasonal variation of solar radiation, affects all parts of the globe, the annual timing of its operation in a particular locality depending broadly upon the latitude. Within the tropics the stimulus operates around the time of the twice-yearly monsoons, whereas north and south of the tropics it operates in the colder months.

Proposition 3, concerning the method of spread of influenza A virus. When persistent virus is reactivated, the carrier host becomes for a short time intensely infectious to his non-immune companions who, if infected, rapidly develop influenza. The carrier host usually suffers no illness from the reactivation. Epidemics consist almost entirely of persons infected by reactivated virus caught from carrier hosts, since the persons infected cannot normally transmit the virus during their illness. Occasional direct transmissions are not excluded.

Proposition 4, concerning the mechanism of antigenic drift. Influenza A virus reactivated from persistent infection usually differs antigenically from its progenitor virus because the immune state of the carrier host induces antigenic drift. Reconstituted particles identical with the progenitor virus cannot readily escape the specific immunity engendered by the carrier's attack of influenza A months or years earlier, so that the carrier can only shed mutant particles from which his non-immune companions unconsciously select the variant(s) fittest (in an evolutionary sense) to survive and maintain the influenza A species.

Proposition 5, concerning the seasonal metamorphosis of influenza A virus. The antigenic character of reactivated virus, although it differs from the progenitor strain, is nevertheless determined by the parent virus. Carrier hosts of the same persistent virus will tend to have produced a similar immune response (the basis of serological identification) and to shed a similar assortment of mutants from which their non-immune companions will select the fittest. Thus last season's prevalent virus may automatically disappear and be replaced next season through the whole area of its prevalence by the successor(s). In some seasons two or more mutants of equally good potential may be produced and co-circulated.

Proposition 6, concerning the phenomenon of antigenic shift. The era of prevalence usually lasts until almost all susceptible persons in the world have been infected and immunized in successive seasonal epidemics. No further epidemic by strains

of the same major serotype can occur until a sufficiency of non-immune persons has been born.

Residues of viruses of earlier major serotypes, latent in older persons, were being reactivated seasonally at low multiplicity, but their chance of pandemic distribution in competition with strains of the prevalent major serotype were negligible. When, however, the current major serotype has completed its era by specifically immunizing virtually the whole world population, this difficulty is removed, and the opportunity is immediately seized. These reactivations of latent residues occurring seasonally in older carrier hosts all over the world must lead to competition between strains of the various major serotypes. The order of precedence must, however, have been long ago determined by evolution, and the rota of major serotype prevalence is conserved.

Proposition 7, concerning epidemics occurring out of season. If one or more carrier hosts, in whom persistent virus is just being reactivated, are rapidly transported to a distant locality where influenza is not seasonable, their non-immune companions in the new locality may catch influenza. The unseasonable outbreak will be limited to those infected contacts, because the virus will promptly become persistent in them, but it may be reactivated locally next season, perhaps only a few months later. If so the strains which they then transmit may be out of step and differ antigenically from the other strains causing influenza in the locality in the same season.

Proposition 8, concerning rapidity of spread of influenza A. The speed with which influenza A appears to travel over the globe reflects the annual movement of the seasonally mediated stimulus recalling virus from persistence in carrier hosts and providing the opportunity for epidemics in non-immune companions of the carrier hosts. The stimulus is dependent on variations in solar radiation, an extra-terrestrial influence unaffected by the rapidity of human travel. The rapidity of influenzal spread was as rapid in previous centuries as it is at present because it does not depend on case-to-case transfer.

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