2

Taxonomy and Comparative Virology of the Influenza Viruses

INTRODUCTION

Viral taxonomy has evolved slowly (and often contentiously) from a time when viruses were identified at the whim of the investigator by place names, names of persons (investigator or patient), sigla, Greco-Latin hybrid names, host of origin, or name of associated disease. Originally studied by pathologists and physicians, viruses were first named for the diseases they caused or the lesions they induced. Yellow fever virus turned its victims yellow with jaundice, and the virus now known as poliovirus destroyed the anterior horn cells or gray (polio) matter of the spinal cord. But the close kinship of polioviruses with coxsackievirus B1, the cause of the epidemic pleurodynia, is not apparent from names derived variously from site of pathogenic lesion and place of original virus isolation (Coxsackie, New York).

In practice, many of the older names of viruses remain in common use, but the importance of a formal, more regularized approach to viral classification has been increasingly recognized by students and practitioners of virology as more basic information has become available about the nature of viruses. Accordingly, the International Committee on Taxonomy of Viruses has devised a generally accepted system for the classification and nomenclature of viruses that serves the needs of comparative virology in formulating regular guidelines for viral nomenclature. Although this nomenclature remains highly diversified and at times inconsistent, retaining many older names, it affirms "an effort. . . toward a latinized nomenclature" (Matthews, 1981) and places primary emphasis on virus structure and replication in viral classification.

E. D. Kilbourne, *Influenza*

TAXONOMY OF INFLUENZA VIRUSES

"Influenza" is the name conferred not by taxonomists or scientists but apparently by its victims in the middle of the 18th century (Creighton, 1891). Whether this Italian word referred to "influenza di freddo" [influence of the cold (Francis, 1959)] or astrological influence (International Committee on Taxonomy of Viruses, 1982) is unclear, but surely the word is appropriate for its potentially devastating effects if not its causation. The viruses causing influenza have been alphabetically named in the chronological order of their isolation and definition as influenza A, B, and C viruses. (Along the way, deficiencies in basic knowledge led to brief consideration of Sendai virus, now classified as parainfluenza virus 1, as influenza D virus.)

As the first viruses recognized to possess in vitro enzymatic activity and to attach to an identifiable substrate, influenza viruses were christened myxo (mucin-reacting) viruses, later changed to orthomyxovirus as the paramyxoviruses were distinguished from and related to them on the basis of evolving knowledge of viral structure and function (see Table 2-1).

Taken literally, the term orthomyxovirus combines the Greek word *orthos* for "straight or correct" and another Greek word, *myxa*, for "mucous," with an Italian form of the Latin *influentia*. Pity the archeologist of the future who has to decipher this designation!

As with no other virus group, the epidemiologic orientation of influenza virus research has led to the acquisition of a vast inventory of virus strains and the need for a variety of prototype strains with which to conduct research. It has long been traditional, therefore, to identify new influenza strains in terms of place of origin, isolate number, and year of isolation, e.g., A/Puerto Rico/8/34, which has become, in laboratory vernacular, simply "PR8." Improved methods of antigenic analysis and determination of viral protein structure, although they have aided enormously in the sorting out of viruses causing human and animal disease, have also blurred the perception of host of origin—e.g., is "swine" influenza virus a human or animal virus?—as a cardinal taxonomic criterion.

Although some antigenic cross reactivity is sporadically demonstrable among them, antigenically distinct subtypes have been defined for 13 hemagglutinins and nine neuraminidases of human, swine, equine, and avian influenza A viruses (Table 2-2). Of these, only H1N1, H2N2, and H3N2 viruses have been isolated from epidemic human infections.

Among avian species various combinations of hemagglutinins and neuraminidases are found, suggesting, along with other evidence, genetic reassortment of the genes for these proteins. In recent years transmission of H3N2 virus, presumably from man to swine, has resulted in combination of this virus with traditional swine (H1N1) virus in Japan to produce hybrid H1N2 virus (Yasuhara et al., 1983).

Because conventional antigenic analyses of influenza viruses identify only their surface antigens, routine methods are obviously incapable of identifying genes for internal or nonstructural proteins and, therefore, can detect only hemagglutinin/neuraminidase antigenic hybrids or reassortants. The application of RNA

Table 2-1. Classification of the Influenza Viruses[a]

Taxonomic status	International name	English vernacular name	Viral envelope antigens[b]
Family	Orthomyxoviridae	Influenza virus group	—
Genus	*Influenza virus*	Influenza virus	—
Type	A	Influenza A virus	—
Subtypes (human viruses)			H1N1 H2N2 H3N2
Strain designation (example)		A/England/1/51 (H1N1)[c] A/Swine/Iowa/15/30 (H1N1)[d]	
Type	B	Influenza B virus	
Subtypes	None		
Strain designation (example)		B/Great Lakes/1/54	—[e]
Type	C[f]	Influenza C virus	
Subtypes	None		
Strain designation (example)		C/Paris/1/67	

[a]Data from Fourth Report of the International Committee on Taxonomy of Viruses (1982) and WHO Memorandum (1980)
[b]H, hemagglutinin, N, neuraminidase
[c]Type/place of isolation/strain number/date of isolation (subtype)
[d]Host of origin is given when original isolate is from a nonhuman host
[e]H and N antigens but no subtypes have been designated
[f]Classification of influenza C virus as a member of the genus *Influenza virus* is provisional

Table 2-2. Subtypes of Hemagglutinin and Neuraminidase Antigens of Influenza A Viruses[a]

Hemagglutinin subtype	Former designation(s)	Neuraminidase subtype	Former designation(s)
H1	H0,H1,Hswl	N1	N1
H2	H2	N2	N2
H3	H3,Heq2,Hav7	N3	Nav2,Nav3
H4	Hav4	N4	Nav4
H5	Hav5	N5	Nav5
H6	Hav6	N6	Nav1
H7	Heq1,Hav1	N7	Neq1
H8	Hav8	N8	Neq2
H9	Hav9	N9	Nav6
H10	Hav2		
H11	Hav3		
H12	Hav10		
H13	—[b]		

[a]WHO Memorandum (1980)
[b]Proposed as new H subtype, avian source (Hinshaw et al., 1982, 1983)

gel electrophoresis, oligonucleotide mapping, and RNA-RNA hybridization techniques have identified such reassortants as A/California/10/78 (H1N1) that clearly have inherited genes from both H1N1 and H3N2 parental human viruses (Young and Palese, 1979; Bean et al., 1980). No provision currently exists for classification of natural reassortants.

Laboratory-produced reassortant viruses (widely used as vaccine strains or reference reagents), if not antigenically hybrid and thus obviously reassortants, bear a postscript "R" after the strain donating the surface antigens. For example, X-31 [laboratory designation (Kilbourne, 1969; Kilbourne et al., 1971)], an early high-yielding vaccine strain widely employed in chemical and immunologic studies of the virus, is properly designated A/Aichi/2/68 (H3N2) (R). This designation, however, without reference to the original papers describing its origin, gives no hint that six of the eight RNAs are derived from PR8 [A/PR/8/34 (H1N1)] (Baez et al., 1980). If antigenically hybrid, reassortants should be identified with respect to the strain of origin of the hemagglutinin and neuraminidase antigens, e.g., A/Bel/42 (H1)-Singapore/1/57 (N2) (R) (WHO Memorandum, 1980).

In short, present taxonomic labels, although useful, do not substitute for precise description as an identification of influenza virus strains in formal scientific communications. No less important is citation of the laboratory of origin. PR8 virus strains from different laboratories may be antigenically different (Paucker, 1960), and Cambridge and Mount Sinai strains of PR8 differ cytopathogenically in human conjunctival cell cultures (J. L. Schulman and E. D. Kilbourne, unpublished data).

The 50-year genealogy of another classical prototype strain, WSN, illustrates not only the hundreds of laboratory passages that it has undergone but the variety of host tissues in which the virus has replicated since leaving the respiratory tract in man in 1933 (see Fig. 6-1). This virus, long used in genetic studies (reviewed by Kilbourne, 1963), defies conventional classification, and, indeed, few working with the virus today know its exact history.

RELATION OF INFLUENZA VIRUSES TO OTHER ENVELOPED VIRUSES WITH RNA GENOMES

A wide variety of viruses have evolved using linear single-stranded RNA as their genetic moiety and host-derived envelopes as their limiting membranes. Such viruses contain virus-coded spikelike projections on their surfaces and share replication characteristics to the extent that pseudotypes—virus particles with mixed envelope antigens—may be formed during mixed infection. However, data in Table 2-3 emphasize critical differences as well as superficial similarities among them. RNA recovered from toga- and coronaviruses is infective, being of positive or message sense (Baltimore, 1971), whereas the strategy of influenza virus replication requires transcription with an endogenous, virus-coded transcriptase of a negative-sense RNA genome to form mRNA. Replication of the Retroviridae is fundamentally different from that of all other enveloped viruses, emphasizing again the superficiality of surface structure as a taxonomic criterion, both literally and figuratively.

Table 2-3. Taxonomic Relationship of Influenza Viruses (Orthomyxoviruses) to Other Enveloped, Single-Stranded RNA Animal Viruses[a]

Genome strategy	Family or group	Genus [example(s)]	Species example (English vernacular name)	Host(s)
Positive sense	Togaviridae	Arbovirus group A	Sindbis virus	Vertebrates, arthropods
		Arbovirus group B	Yellow fever virus	Vertebrates; arthropods
		Rubella virus	Rubella virus	Man
		Mucosal disease virus group	Hog cholera virus	Vertebrates
	Coronaviridae	Coronavirus	Human coronavirus	Man
Negative sense	Paramyxoviridae	Paramyxovirus group	Parainfluenza virus 1 (Sendai)	Man, mouse
		Measles–rinderpest distemper group	Measles virus	Man
		Respiratory syncytial virus group	Respiratory synctial virus	Man
	Orthomyxoviridae	Influenza virus group	Influenza A virus	Vertebrates
	Rhabdoviridae	Rabies virus group	Rabies virus	Vertebrates, arthropods
	Bunyaviridae	Bunyamwera supergroup	Bunyamwera virus	Vertebrates; arthropods
	Arenaviridae	LCM virus group	Lymphocytic choriomeningitis virus	Vertebrates
DNA replication step	Retroviridae	(RNA tumor viruses)	—	Vertebrates

[a]Data derived from Fourth Report of the International Committee on Taxonomy of Viruses (1982)

More closely related structurally are the paramyxoviruses, some of which share with influenza viruses an external virus-coded neuraminidase. Comparison of the influenza and parainfluenza viruses (Table 2-4) reminds one of the original disease-related basis for viral taxonomy in that both of these viruses with hemagglutinating and neuraminidase activity primarily attack respiratory tissue with the production of acute respiratory tract disease. Indeed, homologies of regions of the HA and NA proteins with the HN protein of a parainfluenza virus have been noted (Blumberg et al., 1985). But paramyxoviruses, especially measles virus, can establish persistent infection, a characteristic ascribed to swine influenza virus by Shope (1935–1936) but not yet verified as a mechanism of influenza pathogenesis. The characteristic differences in influenza and parainfluenza viruses with respect to epidemiologically significant antigenic variation is not totally explicable on the basis of differences in their capacity to participate in genetic reassortment.

INFLUENZA VIRUSES AS SEGMENTED GENOME VIRUSES

No system of nomenclature relates the diverse viruses that carry their genetic information in discrete segments. However, viruses possessing this type of genome have the capacity for genetic reassortment with related strains and, therefore, the potential for rapid evolution of essentially new viruses derived from biparental contribution analogous to sexual reproduction. Potential advantages to viruses capable of genetic reassortment include the facilitation of interspecific infection and adaptation to extend viral host range (Kilbourne, 1981). Other advantages are cited in Chapter 6. The taxonomic implications of reassortment

Table 2-4. Comparison of Influenza and Parainfluenza Viruses

Viral property	Influenza viruses	Parainfluenza viruses
Genome	Segmented: eight molecules of linear ss RNA	One molecule of linear ss RNA
Genome strategy	Negative sense	Negative sense
Genetic Reassortment	+	0
Require host cell nuclear functions	+	0
Virion size (diameter)	80–124 nm	150 nm or more
Enveloped, budding virus	+	+
Hemagglutinin and neuraminidase	Two separate glycoproteins[a]	Both functions in single glycoprotein
Epidemiologically significant antigenic variation	+	Unproved
Persistent infection	Unproved	+

[a]One glycoprotein in influenza C virus

Table 2-5. Prototype Segmented Genome Viruses of Vertebrates and Their Host Range[a]

Genus	Subgroup	Prototype	RNA	Segments	Gene products	Host range
Reovirus	Reovirus	Reovirus type I	ds	10	11	Mammals, birds
Reovirus	Orbivirus	Bluetongue virus	ds	10	9	Mammals, insects
Reovirus	Rotavirus	Human rotavirus	ds	11	10–12	Mammals
Birnaviruses[b]	—	Infectious pancreatic necrosis virus	ds	2	6	Fish
Birnaviruses[b]	—	Infectious bursal disease virus	ds	2	6	Chickens
Bunyavirus	—	Bunyamwera virus	ss	3	5	Mammals, insects
Orthomyxovirus	—	Influenza A virus	ss	8	10	Mammals, birds
Arenavirus	—	LCM virus	ss	2	3–4	Mammals

[a]Revised from Kilbourne (1981)

[b]Bisegmented ds RNA animal virus group

are in its potential for the instant creation of new viruses as genes from two or more viruses are mixed in varying proportions, an event now found to have occurred with cocirculating influenza viruses in nature (Young and Palese, 1979; Bean et al., 1980).

Examples of the varied viruses of vertebrates with segmented RNA genomes are listed in Table 2-5, which includes both single- and double-stranded viruses containing two to 11 RNA segments that code for three to 12 gene products. These viruses are widely distributed in nature and infect a variety of hosts, including insects, fish, birds, and mammals. The role of segmented genomes in viral evolution, adaptation, and survival has not yet been demonstrated, but by facilitating genetic interchange, they appear to enhance the evolutionary and adaptive advantage of viruses possessing them (Kilbourne, 1981). Natural reassortment of viruses other than influenza viruses has not been reported.

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