CHAPTER 1

Epidemiologic Concepts and Methods

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1. Introduction

The epidemiology of infectious diseases is concerned with the circumstances under which both infection and disease occur in a population and the factors that influence their frequency, spread, and distribution. This concept distinguishes between infection and disease because the factors that govern their occurrence may be different and because infection without disease is common with many viruses. Infection indicates the multiplication of an agent within the host and is determined largely by factors that govern exposure to the agent and by the susceptibility of the host. Disease represents the host response to infection when it is severe enough to evoke a recognizable pattern of clinical symptoms. The factors that influence the occurrence and severity of this reponse vary with the particular viruses involved and their portal of entry, but the most important determinants for many common infections lie within the host itself. Of these, the age at the time of infection, genetic background, and immune status of the host are the most crucial.

This first chapter deals in a general way with concepts, methods, and control techniques that are explored in detail in individual chapters concerned with specific viruses or groups of viruses. For fuller presentations of the epidemiologic principles, see references 42, 75, 87, 121, 174, 176, and 192.

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2. Definitions and Methods

Incidence is the number of new cases of disease occurring in a unit of time. The incidence *rate* is the number of new cases over the total population at risk. The numerator in this ratio is usually based on the number of *clinical cases* of the disease in question as recognized by physicians and reported to public health departments over the period of a year. The denominator represents the population under surveillance. This is often the total population of the geographic area encompassed by the reporting system. In more intensive studies, the numerator may be defined as the incidence of infection (with or without disease) as determined by viral excretion and/or the appearance of antibody between two points in time. The denominator may be defined as those who are both exposed and susceptible (i.e., lack antibody). These more sophisticated definitions are usually restricted to special investigations in which antibody or viral measurements, or both, are possible.

Prevalence is the number of cases existing at one time. The *prevalence rate* is the number of such cases divided by the population at risk. The time period involved may be 1 year or other fixed period (period prevalence) or a given instant of time (point prevalence). The term *period prevalence* involves both the number of new cases (incidence) and the duration of illness (number of old cases persisting from the previous reporting period). It is used most commonly for chronic diseases.

In serological surveys, *prevalence* represents the presence of an antibody, antigen, chemical marker, or other component in blood samples from a given population at the time of the collection. The *prevalence rate* is the number of sera

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with that component divided by the number of persons whose blood was tested. For viral infections, the prevalence of antibody represents the cumulative infection rate over recent and past years depending on the duration of the antibody. For neutralizing or other long-lasting antibody, it reflects the lifetime or cumulative experience with that agent. If the antibody measured is of short duration such as IgM antibody, then prevalence indicates infection acquired within a recent period.

Descriptive epidemiology deals with the characteristics of the agent, the environment, and the host and with the distribution of the resultant disease in terms of place, season, and secular trends. It is concerned with what the late John R. Paul[174] called "the seed, the soil and the climate." The delineation of these attributes of infection and disease in a population is the "meat" of epidemiology, and this text is largely one of this descriptive nature. The sources of data on which this volume is based are mortality and morbidity reports, field and serological surveys, and special investigations that are described in detail in Chapter 2.

Analytical epidemiology is concerned with planned epidemiologic investigations designed to weigh various risk factors or to evaluate a hypothesis of causation. Two methods of analytical study are commonly employed: the prospective or cohort and the retrospective or case-control study. These are discussed in detail in a recent book on methods in observational epidemiology dealing with both infectious and chronic disease epidemiology.[121] Serological methods are presented in Chapter 2 of this volume and in references 57, 94, and 212.

The prospective method is a means of measuring incidence in a population or a cohort observed over time. In virology, incidence studies permit the direct assessment of the risk of infection or disease, or both, in a defined population group over time in terms of age, sex, socioeconomic level, and other factors. Both the numerator and the denominator are known. In practice, incidence rates of clinical disease are often calculated retrospectively by using data on cases and populations that have been filed away; in virology, total infection rates, with or without clinical illness, can be determined by carrying out virus isolations or serological tests, or both, on materials that have been frozen away and for which data on the population sampled are available. Since such studies are not "prospective" in terms of the observer, calling them "cohort," "longitudinal," or "incidence" studies is more appropriate in a semantic sense. In addition to the direct measurement of risk, this type of investigation avoids the need of selecting controls, because one is merely recording the occurrence of disease or of infection in persons with different characteristics. The disadvantages of incidence studies are that they are expensive because an entire population must be kept under observation and appropriate specimens collected; the lower the incidence of the disease, the larger the denominator requiring observation, and the higher the expense. They are sometimes laborious to conduct and may require much technical help.

Retrospective or case-control studies compare the presence or absence of certain suspected etiologic factors in patients with a certain disease to their occurrence in subjects without this disease. An example is the relationship of smoking to the occurrence of lung cancer. Since both the disease and the characteristic are already present at the time of observation, the data obtained represent prevalence rather than incidence rates. The absolute risk of the disease in persons with different characteristics cannot be measured because no denominators are available. Only the relative prevalence of the disease in persons having the characteristic can be calculated. These are termed "relative risk" or "odds" ratios. The selection and identification of appropriate controls in retrospective studies often pose difficulties because unrecognized biases may be present. In virology, an example of the case-control method would be the evaluation of the etiologic role of a given virus in a certain disease by comparison of the frequency of viral excretion and/or antibody rises in patients having this disease with their frequency in those not having the disease. In evaluating this relationship, it must be remembered that infection without clinical disease is common in viral infections and might be occurring in the control group. Another recent example is comparison of the frequency of elevated viral antibody titers in the sera of patients with certain malignant or chronic diseases with those of age- and sex-matched controls as a clue to causation. Examples of this are the relationship of raised antibody levels of Epstein–Barr virus (EBV) to Burkitt lymphoma and nasopharyngeal cancer as compared to controls, or of measles antibody titers in cases of subacute sclerosing panencephalitis and multiple sclerosis in relation to controls. In general, retrospective or case-control analyses are cheaper, are more quickly performed, and require smaller numbers than incidence studies but measure relative rather than absolute risk.

Traditionally, the existence of a possible causal association between a factor and a disease is usually recognized in a clinical setting, and its statistical significance is determined by comparison with controls using the case-control or retrospective method. If the results indicate the presence of an important association, an incidence study is then set up to evaluate or confirm the observation. Thus, the risk of smoking in lung cancer and that of rubella infection in congenital abnormalities were discovered by case-control methods and confirmed by incidence and cohort analyses. Other retrospective case-control investigations such as those on the relationship between certain blood groups and influenza[81,145] have not been confirmed when tested using incidence data.

Experimental epidemiology utilizes epidemiologic models and is the most elegant and sophisticated approach because all the variables should be subject to control. Unfor-

tunately, animal models may be difficult or impossible to establish in the laboratory, and even if they are established, there is sometimes the question of the applicability of the results to the human host. Theoretically, the ideal way would be the employment of volunteers. In the past, human subjects have participated in studies of yellow fever, malaria, hepatitis, infectious mononucleosis, acute respiratory infections, measles, rubella, and even syphilis. Such investigations involved important technical, medical, ethical, and moral issues. On the technical level, there is the question of the susceptibility of the volunteer to the disease under study; i.e., volunteer adults may already be immune as a consequence of childhood infection. Second, the host response to many infections may result in disease in only a small percentage of those exposed or even of those infected, thus requiring a large volunteer group. Medically, there is concern for the seriousness of the disease produced and for the possibility, however remote, of permanent disability or even death. Finally, the moral and ethical right to use human subjects in any medical experimentation is under debate. In today's climate, experimental studies in volunteers are subject to very strict control, and work being supported by government, foundation, or institutional funds must be scrupulously reviewed by a committee of professional and sometimes of lay and religious representatives. This peer group is required to weigh the benefits of the experiment against the risks involved and to ensure that the experimental subjects are fully aware of all possible consequences before signing a statement of "informed consent."

Serological epidemiology is a term applied to the systematic testing of blood specimens from a defined sample of a healthy population for the presence or level of various components. These include antigens, antibodies, proteins, biochemical and genetic markers, and other biological characteristics (see Chapter 2 and references 57, 94, 175, and 212).

3. Epidemics

An *epidemic* or outbreak of disease is said to exist when the number of cases is in excess of the expected number for that population based on past experience. This determination obviously requires a knowledge of the number of both current and past cases. The definition of "excess" is an arbitrary one and depends on the concentration of cases in any given place, time period, or population group. The occurrence of a large number of cases, compressed in time, as when a new influenza strain is introduced, is readily identified as an "epidemic." Indeed, for influenza, a more sophisticated index has been set up by the National Centers for Disease Control in the United States by which an expected threshold of deaths from influenza and pneumonia in 122 cities has been established based on a 5-year average. When this threshold is exceeded, an influenza outbreak is said to exist. In contrast, even a few cases of encephalitis over a summer may constitute an "outbreak" in areas where no cases previously existed. When several continents are involved, a disease is said to be "pandemic." The current global outreach of the acquired immune deficiency (AIDS) represents such a pandemic.

Chronic diseases pose more difficult problems in definition because their scale of occurrence must be viewed over years rather than months or weeks. In such a perspective, we do have current "epidemics" of chronic illnesses such as coronary artery disease, lung cancer, and intravenous drug abuse. The use of cocaine, especially in its free-base form or "crack," is posing an epidemic threat in the United States. The key words are "an unusual increase in the expected number of cases," irrespective of whether the time period involved is short or long.

Three essential requirements for an outbreak of viral disease are the presence of an infected host, an adequate number of susceptibles, and an effective method of contact and transmission between them. If the agent is not endemic within the community, then the introduction of an infected person, animal, insect, or other vector of transmission is needed to initiate an outbreak. This is particularly important in a remote island or isolated population group, where a virus disappears after no more persons remain susceptible, if persistent viral excretion does not occur to permit infection of newborns. Rubella, for example, disappeared from Barbados for 10 years despite an accumulation in the number of susceptibles to a level representing about 60% of the population and despite the existence of a large tourist trade.[74] In an isolated Indian tribe in Brazil, antibodies to respiratory-transmitted viruses including measles, influenza, and parainfluenza were essentially absent from the entire tribe.[21] The introduction of more susceptibles or of more infected persons may tip this balance. However, antibodies to viruses characterized by persistent or recurrent viral excretion, such as herpes viruses and adenoviruses, have been present in every population thus far tested, no matter how remote or isolated.[21]

The cumulative number of persons immune to a given disease within a community has been termed the *herd immunity* level. If this level is sufficiently high, then the occurrence of an outbreak has been regarded as highly unlikely. For highly communicable infections such as rubella or measles, the level of herd immunity must be of the order of 95% or higher to be effective. For example, in an open college community, a preexisting herd immunity level to rubella of 75% failed to prevent an outbreak of this disease.[79] Indeed, the rubella infection rate of 64% among those completely susceptible (i.e., without detectable antibody) was even

higher than the 45% infection rate in the same community for a new influenza strain to which the entire population was susceptible.[79] A rubella outbreak has even occurred among military recruits in the presence of a 95% level of herd immunity: 100% of the susceptibles were infected.[109] The spread of infection is apparently so efficient under these circumstances of close and prolonged contact that a high level of herd immunity does not deter its progress. Another possibility is that reinfection of partially immune persons results in pharyngeal excretion and further spread of virus to susceptible persons.

For smallpox, the induction of herd immunity by vaccination has resulted in the complete global eradication of the disease through the efforts of the World Health Organization. The last case occurred in Somalia on October 26, 1977.[214] No new natural cases have been reported for at least 10 years since then, although laboratory infections have occurred and remain a hazard to laboratory personnel. Continued surveillance will be needed to assure eradication, especially from sources such as the laboratory or biological warfare or animal reservoirs of smallpox-related viruses. The spread of monkeypox to susceptible persons in endemic areas who have been born since smallpox vaccination of the general population was discontinued in 1981 deserves continued attention.

Mathematical models have been constructed to fit the epidemic spread of certain infectious diseases or as a basis for immunization programs.[1,6,10,55] For diseases in which most infections are clinically expressed, the immunity is good, the means of transmission is limited to one or two routes, the mixture of susceptibles and immunes is homogeneous and equally distributed, and where the age at the time at which infection occurs is figured in the calculations, then such mathematical models may be useful in planning control measures. They require the input of a good mathematician and good epidemiologist, both of whom understand the dynamic interplay of these various factors. Even then, the model must be based on a particular population group with consideration to their socioeconomic status, population mixing, vaccination programs, and behavioral characteristics. The model must then be tested over time in that population group against the actual number of cases reported in a good surveillance system. A reasonably accurate prediction of actual events has been achieved in a model developed for rubella and measles vaccination programs by Anderson and May.[6] But in other situations, where there are many inapparent infections or the disease results from reactivated rather than primary infection or in which the agent is intermittently excreted in the infected host or intermittently present in some environmental or arthropod vector, then the events leading to infection and disease are so complex and variable that a mathematical model is difficult to construct. The limitations for such models and recommendations for their improvement based on better data has been well reviewed by Singer[193] using malaria as the example.

4. Investigation of an Epidemic

The investigation of an epidemic involves a sequence of steps summarized in Table 1. They do not necessarily represent the appropriate order of execution. It may not be possible to establish a definitive diagnosis early, so a rather specific, simple working definition should be established using key epidemiologic and clinical features as a case-finding device. This definition can be expanded and made more sensitive later, when laboratory studies are possible. Control measures should be instituted as soon as the means of spread is reasonably established. Common source outbreaks of viral infections from water, food, milk, or environmental sources are not nearly as common as with bacterial infections. However, they do occur. Some examples include spread of adenoviruses by eye tonometers in eye clinics or via swimming pools, of hepatitis A by public water supplies or by seafood, of hepatitis B by virus-contaminated yellow fever vaccines, or of enteroviruses by fecally contaminated foodstuffs or milk. Most common viral epidemics are respiratory or arthropod-borne, and more recently, spread of several types of viral infections in hospital settings has been recognized.

The worldwide epidemic of AIDS involving the spread of the human immunodeficiency virus (HIV) is a cause of major concern. In the United States alone there are over 1.5 million infected with the virus as of mid-1988, of whom some 60,000 have already developed clinical AIDS. It is estimated that at least 50–80% of those infected will develop AIDS, AIDS-related complex, or some other manifestation

Table 1. Epidemic Investigation

1. *Define the problem.* Diagnosis? Is it an epidemic?
2. *Appraise existing data*
   Time: data (and hour) of onset; make epidemic curve
   Place: spot map of cases; home, work, and recreational places; special meetings
   Person: age, sex, occupation, ethnic groups
   Incidence rates: infection, cases, deaths
   Possible means of transmission
   Seek common denominator and unusual exceptions
3. *Formulate hypothesis.* Source of infection, method of spread, possible control
4. *Test the hypothesis.* Search for added cases; evaluation; laboratory investigation
5. *Conclusions and practical application.* Long-term surveillance and prevention

in the 4- to 7-year incubation period. There is no evidence of a decrease in the doubling time of those infected or diseased, although transfusion-transmitted infection has been almost eliminated by the use of screening tests for antibody to HIV; some infected donors will escape detection because they are in the incubation period and have not yet, or perhaps never will, develop antibody. The mortality is at least 70%–90% for AIDS because of the current lack of effective therapy. Without the ability to prevent the development of clinical illness among the infected by antiviral therapy or to produce a vaccine because of the antigenic variability of the isolates, a serious global threat to human life exists. In developed countries, the spread of the virus from homosexuals and intravenous drug abusers into the community via heterosexual transmission and from mother to infant via perinatal means poses the greatest risk. In Africa, the widespread and equal occurrence of infected males and females and the presence of related viruses in nonhuman primates are the major concerns. Intensive efforts to prevent further spread from those infected and to prevent susceptibles from entering high-risk activities by educational and regulatory means are of utmost importance. The epidemiology and control of retroviruses are discussed in Chapter 21. The steps to investigate epidemics are presented in greater detail in Chapter 1 of the companion volume to this, *Bacterial Infections of Humans: Epidemiology and Control,* as well as in a recent book, *Methods in Observational Epidemiology.*[121]

5. The Agent

This section is concerned primarily with those general properties of viruses that are important to an understanding of their epidemiology and not with their basic clinical chemistry, morphology, genetics, or multiplication. These aspects are dealt with in various microbiology and virology textbooks.[15,47,85,89,90,112,131,139,142,199,201,208,218]

The chief characteristics of viruses that are of importance in the production of infection in man are (1) factors that promote efficient transmission within the environment; (2) the ability to enter one or more portals in man; (3) the capacity for attachment to, entry into, and multiplication within a wide variety of host cells; (4) the excretion of infectious particles into the environment; and (5) a means of developing alternative mechanisms of survival in the face of antibody, cell-mediated immunity, chemotherapeutic agents, interferon, or other hostile elements. Survival of the virus might be achieved through mutation, recombination, basic properties of resistance, or the availability of alternative biochemical pathways.

Intensive studies of the various parts of the viral genome responsible for particular functions, the dynamics of infection at the cellular level, and the specificity and complexity of the immune response in the susceptible host are now being carried out for many viral infections. A recent virology book edited by Fields *et al.*[85] is an excellent source of these advances and their application to clinical virology. For example, it is now clear that a minor change in a single gene of a particular virus, such as the reovirus, or even in a single nucleotide or amino acid, such as in rabies virus, may have a profound effect on the pathogenicity of the agent and the pattern of clinical disease that develops.[166,189] Similarly, subtle changes in the immune system can alter the host's response to the same virus.[166,167]

The spread of viruses depends on (1) the stability of the virus within the physical environment required for its transmission, including resistance to high or low temperatures, desiccation, or ultraviolet; (2) the amount of virus expelled into the proper vehicle of transmission; (3) the virulence and infectivity of the agent; and (4) the availability of the proper vector or medium for its spread.

After entry through an appropriate portal, the virus must escape from ciliary activities, macrophages, and other primary defense mechanisms during its sojourn to the target cell, find appropriate receptors on the cell surface for its attachment, and be able to penetrate and multiply within the cell. The steps then include initiation of transcription of messenger ribonucleic acid (mRNA), translation of early proteins, replication of viral nucleic acids, transcription of mRNA, translation of late proteins, assembly of virions, and then viral release.[84] These aspects fall into the province of basic virology and are not discussed in detail here. What is important in pathogenesis is the efficiency of spread from cell to cell, either by direct involvement of contiguous cells or by transport via body fluids to other susceptible cells; the number of cells infected; and the consequences of viral multiplication on the cell itself and on the organism as a whole. The long-term survival of a virus in human populations depends on its ability to establish a chronic infection without cell death or on an effective method of viral release into the environment in a manner ensuring its transport to a susceptible host, or on a highly adaptive system for biological adversity. The prime example of adaptability among animal viruses is influenza A. Without its propensity for antigenic variation, it would probably behave like measles or rubella viruses and be dependent for survival on the temporal accumulation of new susceptibles.

6. The Environment

The external environment exerts its influences on the agent itself, on the manner of its spread, and on the nature of the host response to infection. Although viruses survive or die within defined ranges of certain physical factors such as temperature and humidity, there is much variability from one

viral group to another. A simple environmental factor such as cold may have different effects on the survival of different viruses and on their ability to multiply within cells. Although environmental characteristics play an important role in the survival of a virus, they are probably of much greater significance in their influence on the routes of transmission and on the behavior patterns of the host.

For infections that require an insect vector, such as the arboviruses, the environment exerts an obvious role in restricting the occurrence of infection and disease to those areas that have the proper temperature, humidity, vegetation, amplifying animal hosts, and other features necessary for the insect involved. For viral diseases potentially transmitted by water, such as hepatitis A virus, non-A, non-B, A-like hepatitis virus, and Norwalk agent, a warm environment attended by poor sanitation and fecal contamination clearly enhances the degree of exposure and the efficiency of transmission.

Perhaps the most crucial effect of climate on common viral diseases is exerted on the social behavior of the host. In tropical settings and in the summer season in temperate climates, the opportunity for transmission of gastrointestinal diseases is increased through contact with water, as in swimming in and drinking from the polluted areas. Warm weather also brings closer contact with dogs and other animal sources of rabies and with insect vectors of arboviruses. In winter, people huddle together indoors, promoting the transmission of airborne and droplet infections. This spread is amplified by the opening of schools and colleges. In addition, the environment within most houses and buildings tends to be hot and dry, which impairs the protective mechanisms of human mucous surfaces and may permit easier entry and attachment of certain respiratory viruses.

Just as winter clearly brings with it an increase in viral respiratory illnesses, heavy rains and the monsoon similarly influence these same diseases in tropical settings. Indeed, the incidence of common upper respiratory diseases in college students was as high in the warm climate at the University of the Philippines as in the intemperate winters at the University of Wisconsin.[71,72] Viruses that cause respiratory infections in children have also been found to be active in all climates around the world.[40] Community studies in India,[169] Trinidad,[20] and Panama[153] have indicated a high morbidity from influenza and other respiratory diseases in tropical settings. As in temperate climates, factors that tend to aggregate people inside, such as heavy rainfall or schooling, also coincide with the highest incidence of respiratory-transmitted infections in the tropics.[72,153]

7. The Host

The factors that influence infection involve primarily exposure to the infectious agent and the susceptibility of the host. The opportunity for a susceptible host to come in contact with a source of infection depends on the means of transmission. Respiratory-transmitted agents are usually general in their exposure; those transmitted by gastrointestinal routes are related to exposure to food or water and to the hygienic and socioeconomic level of the host; those that depend on arthropod-borne transmission involve persons in special settings or special occupational exposures. Others, such as sexually transmitted agents, require specific behavioral acts of the host; still others require specialized exposures such as transfusions, rabid animals, or specialized environments. The factors that influence infection are therefore mostly extrinsic to the host, but not all fully exposed persons will develop infection, as manifested by the appearance of antibody and the isolation and/or demonstration of the causative agent. The agent factors affecting the outcome of its encounter with the host include the dose, infectivity, and virulence of the virus and the number of surviving infectious particles that enter an appropriate portal and find viral receptors on susceptible cells. Host factors include the vigor of the primary defense system, such as cilia, mucus, and nonspecific viral inhibitors, the genetic susceptibility to the virus, and the presence or absence of antibody and cell-mediated immunity.

Those factors that determine whether clinical illness will develop in a person already infected depend in part on the dosage, virulence, and portal of entry of the agent, but more important, they depend on certain intrinsic properties of the host. Some of these characteristics are listed in Table 2. Age at the time of infection is a critical host factor and influences whether clinical illness develops following infection with such agents as Epstein–Barr virus, hepatitis viruses, and poliomyelitis viruses. In general, the probability that clinical illness will develop increases as the age at the time of infection increases; in a similar fashion, the severity of the clinical response also increases with age at the time of illness. The nature of the immune response to a virus can be either beneficial to the host in limiting the infection or detrimental if the clinical disease is caused by certain immunopathological consequences of infection such as immune complexes or autoimmune mechanisms. The vigor of the humoral and cell-mediated immune responses may also determine when a virus becomes persistent or is eradicated from the body.

The severity of the clinical response to viral infections is greatly enhanced when the immune system is compromised as a result of an inherited or acquired immunodeficiency, by immunosuppressive drug therapy as in renal transplant patients, or by infection and destruction of key lymphocytes involved in cell-mediated immunity such as the elimination of susceptible T_4 (helper) lymphocytes by HIV. Certain host attributes also affect the occurrence or severity of certain infections: smoking increases the severity of acute respirato-

Table 2. Factors That Influence the Clinical Host Response

1. Dosage, virulence, and portal of entry of the agent
2. Age at the time of infection
3. Preexisting level of immunity
4. Nature and vigor of the immune response
5. Genetic factors controlling the immune response, the presence of receptor sites, and cell-to-cell spread
6. Nutritional status of the host
7. Preexisting disease
8. Personal habits: smoking, alcohol, exercise, drugs
9. Double infection or bacterial complications
10. Psychological factors (e.g., motivation, emotional crises, attitudes toward illness)

ry infections, as does the presence of maternal antibody in respiratory syncytial viral infections. Alcohol appears to increase the risk of chronicity for certain hepatitis viruses, and exercise predisposes to the development of paralytic poliomyelitis in the exercised limb. Psychosocial factors manifested by increased motivation toward a career, overachieving fathers, and poor academic performance have been shown to increase the frequency of clinical infectious mononucleosis as well as its severity in cadets at the West Point Military Academy infected with Epstein–Barr virus.[70,119] The ability to identify which persons are susceptible at the time of exposure and those who are infected, the frequency of clinical disease among the infected, and the availability of psychosocial data at the start of the study permitted the delineation of factors that would have been obscured if just exposure and disease had been considered.

Nutrition and genetic susceptibility are probably of importance in tipping the scale toward clinical illness, but few studies have been done to measure this. The determinants of clinical expression among the infected have been termed "clinical illness promotion factors" and have recently been reviewed.[70] Our knowledge of the actual cellular and molecular basis for these events is meager, but new virological and immunologic techniques are making rapid advances in our understanding both in humans and in animal models.[150–152,184,189]

8. Routes of Transmission

The major routes of transmission of viral infections are listed in Table 3. Many viruses have several alternate routes, thus enhancing the chance of infection. The sequence of events in transmission involves release of the virus from the cell, exit from the body, transport through the environment in a viable form, and appropriate entry into a susceptible host.

Some viruses are released from cells at the end of the cycle of multiplication. Others do not complete this cycle (incomplete viruses), and some do not effect efficient escape (cell-bound viruses). Many viruses are released from cells by budding, acquiring a lipoprotein coat or envelope as they go through the cell membrane; these include herpesviruses, togaviruses, myxoviruses, paramyxoviruses, and coronaviruses. Nonenveloped viruses not released by budding are the adenoviruses, parvoviruses, poxviruses, picornaviruses, and reoviruses. Some of these latter are released by cell lysis. Once released, viruses find their way to new hosts via one or more portals such as the respiratory tract (e.g., influenza), skin (varicella, smallpox), blood (hepatitis viruses via blood transfusion, arboviruses via mosquitos), gastrointestinal tract (enteroviruses), genital tract (herpes simplex type 2), urine [cytomegalovirus (CMV)], and placenta (rubella, CMV). A more detailed presentation of these major routes of spread is now given.

8.1. Respiratory

The respiratory route is probably the most important method of spread for most common viral diseases of man and is the least subject to effective environmental control. For influenza virus, the degree of transmissibility varies from one strain to another and seems to be independent of other attributes of the virus. Schulman[187] has compared the features of a strain with high transmissibility (Jap 305) and one with low transmissibility (Ao/NWS) in an experimental mouse model. The virus titer in the lung was similar for both strains, but the virus content in the bronchial secretion was low for the Ao/NWS strain compared to the Jap 305 strain. This higher degree of release into the respiratory portal of exit resulted in detectable virus in the air surrounding mice infected by the Jap 305 but not those infected by the Ao/NWS strain. Once an aerosol was created, the stability of both strains was similar. Protein analysis also revealed differences in the neuraminidase of the two strains; this component is associated with dissociation of viruses from the cell and thus perhaps with its transmissibility. However, high transmissibility did not go along with transfer of the gene for neuraminidase, so it was concluded that other factors were also involved in the efficacy of spread.

Other aspects that affect the transmission of respiratory viruses are the intensity and method of propulsion of discharges from the mouth and nose, the size of the aerosol droplets created, and the resistance of the airborne virus to desiccation. Much work has been done by Knight[126] and his group on the transmission of respiratory viruses. At one extreme is the direct transmission of infection via personal contact such as kissing, touching of contaminated objects

Table 3. Transmission of Viral Infections

Routes of exit	Routes of transmission	Example[a]	Factors	Routes of entry
Respiratory	Bite	Rabies	Animal	Skin
	Saliva	EBV	Kissing	Mouth
			Prechewed food, infants	
		HBV	Dental work	
		?HIV	Sexual	
	Aerosol	Influenza, measles	Cough, sneeze	Respiratory
	Oropharynx to hands, surfaces	HSV, RSV, rhinovirus	Fomites	Oropharynx
Gastrointestinal	Stool to hands	Enteroviruses	Poor hygiene	Oropharynx
	Stool to water, milk food	HAV, rhinoviruses	Seafood, water, etc.	Mouth
		HAV, non-A, non-B, A-like		
	Thermometer	HAV	Nurses	Rectal
Skin	Air	Pox viruses	Vesicles	Respiratory
	Skin to skin	Mulluscum contagiosa warts	Abrasions	Abraded skin
Blood	Mosquitoes	Arboviruses	Extrinsic Incubation period	Skin
	Ticks	Group B togaviruses	Transovarial transmission	Skin
	Transfusions of blood and its products	HIV, HBV, non-A, non-B, CMV, EBV	Carrier in plasma or lymphs	Skin
	Needles for injection	HIV, HBV, non-A, non-B	Drug addicts, tattooing	Skin
Urine	Rarely transmitted	CMV, measles, mumps, rubella	Unknown	Unknown
Genital	Cervix	HSV, CMV, HBV, HIV, rubella	Sexual, perinatal	Genital
	Semen	CMV, HBV, HIV	Heterosexual	Genital
			Homosexual	Rectal
Placental	Vertical to fetus	CMV, HBV, HIV, rubella	Infection in pregnancy	Blood
Eye	Tonometer	Adenovirus	Glaucoma test	Eye
	Corneal transplant	Rabies, Creutzfeldt–Jakob disease	Surgery	

[a]CMV, cytomegalovirus; EBV, Epstein–Barr virus; HAV, hepatitis A virus; HBV, hepatitis B virus; HIV, human immunodeficiency virus; RSV, respiratory syncytial virus.

(hands, handkerchiefs, soft drink bottles), and direct impingement of large droplets produced by coughing or sneezing. This last method is regarded as a form of personal contact because of the short range of the heavy droplets formed. Sneezing and coughing also create aerosols varying in size from about 1 to more than 20 μm that permit transmission of infection at a distance. The dispersion of an aerosol depends on wind currents and on particle size. In still air, a spherical particle of unit density of 100-μm diameter requires 10 sec to fall the height of the average room (3 m), 40-μm particles require 1 min, 20-μm particles 4 min, and 10-μm particles 17 min. This means that particles under 10 μm have a relatively long circulation time in the ordinary room. Once initiated, particles 6 μm or more in diameter are usually trapped in the nose, whereas those 0.6–6.0 μm in diameter are deposited on sites along the upper and lower respiratory tract.

Hygroscopic particles of 1.5-μm diameter discharged in large numbers by coughing or sneezing lose moisture and shrink in ambient air but regain their original dimensions from the saturated air in the respiratory tract. The site of disposition of an aerosol containing virus particles does not necessarily represent the level in the respiratory tree where the greatest number of susceptible cells exist for that agent. Quantitative studies have indicated that with four different respiratory viruses, the number of viral particles necessary to produce infection in the respiratory tract is relatively small. With adenoviruses, for example, it is on the order of seven virions. The lower infective dose required for nasal implantation of rhinoviruses and coxsackievirus indicates that this route, perhaps by personal contact, leads to their effective

transmission.[126] The high concentrations of rhinovirus particles on fingers, hands, and hard surfaces as opposed to the lower concentrations found in aerosols suggest that infection via hands may be an important route of spread. This is supported by the frequent inadvertent contact of hands with the nose or eyes.[104] If the importance of this mechanism is confirmed, frequent hand washing may help control the spread of the common cold, as may certain chemicals impregnated into cleaning tissues. Hands and other fomites are also important in the transmission of respiratory syncytial virus.[101]

The size and number of viral particles in sneezes and coughs have varied from study to study depending on the methodology employed. In one study, 1,940,000 particles were present in sneezes and 90,765 in coughing, a ratio of 2.14 : 1.[93] Despite the high level of particles, the recovery of Coxsackie A21 virus itself was more frequent from coughs than from sneezes.[126] Many questions on the mechanics of transmission of respiratory viruses remain unanswered, and any generalizations are premature, but the methodology to answer some of these is becoming available.

Aerosolization of certain viral agents may occur from suction devices and from catheters in intensive care units and from blood products in dialysis units. These include not only respiratory and intestinal agents but also blood-associated agents such as hepatitis viruses and potentially lymphocyte-associated viruses such as CMV, EBV, and HIV.

8.2. Gastrointestinal

Transmission by the oral–fecal route is probably the second most frequent means of spread of common viral infections, and the gastrointestinal tract is the second great portal of entry of infection. Viruses can directly infect susceptible cells of the oropharynx, but to induce intestinal infection, virus-containing material must be swallowed, successfully resist the hydrochloric acid in the stomach and the bile acids in the duodenum, and progress to susceptible cells in the intestine. These cells may be the epithelial cells in the intestinal mucosa or in the intestinal lymphatics, as with adenoviruses. Viruses with envelopes do not normally survive exposure to these acids, salts, and enzymes in the gut. The major enteric viruses are rotaviruses, Norwalk agent, poliomyelitis, echo, Coxsackie, and hepatitis and non-A, non-B A-like viruses. It is known that under conditions of close and prolonged contact, hepatitis B virus and non-A, non-B B-like hepatitis may also be transmitted in this way. Multiplication and excretion in the intestinal tract also occur with adenoviruses and reoviruses, but this route of transmission is not usually of epidemiologic importance. The rhinoviruses are acid labile and do not survive passage through the stomach. Unlike the respiratory viruses, the enteroviruses rarely produce evidence of local disease as a consequence of their multiplication in cells lining that area. Thus, diarrhea, vomiting, and abdominal pain are highly unusual features of infection with these agents. Instead, their major target organs and the site of major symptomatology are at a distance: hepatitis viruses in the liver and enteroviruses in the central nervous system and skin. The HIV virus may be introduced into the rectum during passive anal intercourse and enter the blood through abrasions in the mucous membrane.

Viruses excreted via the gastrointestinal tract must successfully infect other susceptible persons via the oral–intestinal route through fecally contaminated hands, food, water, milk, flies, thermometers, or other vehicles. Viruses spread via these routes are subject to much greater environmental control than are agents transmitted by the respiratory route. Thus, good personal hygiene, especially washing of hands after defecation, proper cleanliness and cooking of food, pasteurization of milk, good waste disposal, and purification of drinking water supplies are effective preventive measures. Hepatitis A virus (HAV) and non-A, non-B, A-type virus are stable viruses in water and, when present in sufficient dosage, may not be inactivated by ordinary levels of chlorine. Furthermore, HAV, at least, can persist in oysters and clams over long periods. This is especially hazardous because these foods are so often eaten without having been cooked. Hepatitis viruses and the enteroviruses also flourish in certain institutional settings (mental hospitals, institutions for retarded children, some prisons) and in countries where personal hygiene is lacking or difficult to practice or where poor environmental control is present. Since some enteroviruses may also multiply in the respiratory tract and be transmitted by the respiratory route, this alternate pathway is of epidemiologic importance even in the face of good personal and environmental hygiene.

8.3. Skin

Skin is the third important area for the entry and exit of viral infections. Although penetration of the intact skin is an unlikely mechanism of infection, the introduction of virus particles via a bite, as with rabies, or via a mosquito, as with the arboviruses, or via a needle or blood transfusion, as with all types of hepatitis viruses and HIV makes this route a very important one. Both CMV and EBV may also be transmitted through blood transfusions. The abraded skin may serve as the entry point of human papovavirus, which causes warts, of hepatitis B virus, and probably for the agent of kuru.

The skin serves as a portal of exit only for those viruses that produce skin vesicles or pox lesions that release infectious particles on rupture. These include herpes simplex, smallpox, varicella–zoster, and vaccinia viruses. The vi-

ruses of certain maculopapular exanthems may also be present in the skin, as in rubella, but this does not seem to be an important avenue of escape, since vesicles are not formed and skin involvement occurs late in the disease, at a time when the virus may be bound by antibody; indeed, the antigen–antibody complex may be responsible for the rash itself.

8.4. Genital

The genital tract serves as a portal of infection for both heterosexual and homosexual partners during sexual activity and is a source of infection as the fetus passes down the birth canal. Herpes simplex, types 1 and 2, CMV, HBV, HIV, and rubella virus are present in cervical secretions and can infect infants during delivery or shortly thereafter; CMV, HBV, and HIV are present in the semen and can be transmitted during either heterosexual or homosexual intercourse.[23,39,116,132,211,219] Long-term asymptomatic cervical or semen carrier states exist and make recognition and control difficult. Passive anal intercourse is an important method of spread of CMV and HIV infections. Genital herpes infections and papilloma virus, type 19, have been implicated in cervical cancer and are discussed in Chapter 26.

8.5. Intrauterine or Transplacental

Viruses may infect the fetus either by direct contact via the birth canal as discussed in Section 8.4 or by hematogenous spread via the placenta to the fetus within the uterus. Viruses that produce intrauterine infection include CMV, hepatitis B, herpes simplex, rubella, varicella viruses, and HIV; CMV and rubella viruses are the most common congenital infections in that order, with congenital rubella decreasing with increasing vaccination. Infection of the fetus may result in no symptoms, in abortion and stillbirth, in developmental abnormalities, in persistent postnatal infection, and in some later manifestations. A fine book[180] and a recent chapter[7] now deal with infectious diseases of the fetus and newborn infant.

8.6. Genitourinary

Although viruses such as CMV and measles are excreted in the urine, this portal of exit has not been established as being of epidemiologic or clinical importance. Considering the wide variety of viruses that can multiply in human kidney tissue cultures *in vitro*, it is surprising that renal infections in man from these viruses are virtually nonexistent or at least are nonrecognized. It seems possible that viruses may play a role in immune complex nephritis in man as they do in experimental animal models, but to date this has not been clearly demonstrated, nor has it been reflected in abnormally high viral antibody levels in such patients.[210] Adenovirus types 11 and 21 have been implicated as the cause of hemorrhagic cystitis in children (see Chapter 3).

8.7. Personal Contact

Direct transfer of infected discharges from the respiratory or gastrointestinal tract to a susceptible person is often included under "transmission by personal contact." Many viruses regarded as "respiratory or airborne" in spread may in fact be more direct in their transmission mechanism, as has been previously mentioned for the rhinoviruses[100,104] and respiratory syncytial virus.[101] On the other hand, person-to-person spread of HIV has not been shown in serological follow-up of families with an index case of AIDS.

8.8. Water and Food

Outbreaks of viral hepatitis have occurred from sewage-contaminated water, as in the large outbreak in New Delhi, India, in 1956,[147] or from seafood obtained from fecally contaminated waters, as shown in outbreaks associated with oysters in the United States and in Sweden[144] and with clams in New Jersey.[51] Milk and water have also served as vehicles of transmission of hepatitis, Norwalk agent, and poliomyelitis viruses. Summer outbreaks of adenovirus type 3 infections have occurred in association with swimming pools.[14]

8.9. Arthropod-Borne

Mosquitos, flies, ticks, and other insects may transmit viral infections. One kind of transmission is a passive type, simply involving survival of the virus in or on the insect that has picked it up from skin lesions or the blood. This type requires neither incubation time in the insect vector nor any specificity for either the arthropod host or the virus. Poliomyelitis and possibly hepatitis viruses may be carried in this way. On the other hand, some viruses require multiplication in the insect vector. In this instance, virus acquired from the blood of the human or animal host during viremia requires a period of multiplication within the arthropod vector before it is infectious, and there is a high degree of vector–virus–host specificity. An example of this is the transmission of yellow fever virus by *Aedes aegypti* mosquitos. The details of arthropod transmission are described in more detail in Chapter 5.

9. Pathogenesis

Since each chapter on specific viruses deals with the subject of pathogenesis, this discussion is limited to a general

consideration of infections involving certain local or systemic features. Good general presentations can be found in other books.[42,150,151,162–166]

9.1. Respiratory

Infectious particles may be implanted directly on nasal surfaces from contaminated hands or from large droplets or may reach the lower respiratory passage from aerosols. Since man continually samples the environmental air about 20 times a minute in breathing, it is no wonder that exposure to and infection with respiratory viruses are common indeed. Furthermore, only a small number of infectious particles need to be implanted in appropriate areas to induce infection. This is on the order of three particles for influenza A by aerosol, six for Coxsackie A21 by intranasal implantation, and seven for adenovirus 4 by aerosol.[126] In general, aerosol particles 3 μm in size reach the alveolus, and those 6 μm or greater are retained in the upper respiratory tract. The mucociliary epithelium transports particles up from the lung or down from the nasal mucosa.[151] To reach susceptible cells, viruses must pass through the mucus film and make physical contact with the cell receptors. The mucus contains mucopolysaccharide and other inhibitors, such as specific immunoglobulin A (IgA) antibody in previously exposed persons. Influenza virus is assisted in its spread by its own neuraminidase, which hydrolyzes the polysaccharides of the inhibitors; the virus attaches to cell receptors by means of surface hemagglutinin spikes. In the alveolus, small aerosol particles are ingested by macrophages, and some viruses are digested and degraded by these cells; other viruses are even capable of multiplication within macrophages themselves.

Most respiratory viruses produce illness through the direct consequences of local multiplication. Necrosis and lysis occur with desquamation of the respiratory epithelium.[47] Constitutional symptoms may then result from breakdown products of dying cells that are absorbed into the bloodstream; fever is produced by the liberation of endogenous pyrogen resulting from viral action on polymorphonuclear leukocytes. This sequence of events may be modified or altered by interferon production in infected cells, by the appearance or preexistence of secretory or local antibody, or by the presence of preexisting or produced humoral antibody. If humoral antibody is present in the absence of local antibody, then a more severe reaction may occur, possibly through antigen–antibody deposition on the cell membrane. The mechanism of this is not clear, but the phenomenon has been observed in infants with passively acquired maternal respiratory syncytial antibody who subsequently develop an infection with this virus. It has also been seen following parenteral administration of an inactivated vaccine that produces humoral antibody but little or no local antibody, such as experimental respiratory syncytial and early measles vaccines when followed by natural or purposeful exposure to live virus.[41]

The multiplication and effect of respiratory viruses such as influenza virus, parainfluenza virus, rhinoviruses, and respiratory syncytial virus are generally limited to the respiratory tract. Influenza virus has been detected in the blood only rarely[52] but has been isolated from the spleen, lymph nodes, tonsils, liver, kidney, and heart in fatal cases of Asian influenza pneumonia.[170] Systemic spread of this type appears to be unusual and associated with overwhelming viral infection.[204] More examples may come to light with more widespread use of immunosuppressive drugs. Adenoviruses and the enteroviruses multiply both in the respiratory tract and in the gut; viremia and secondary multiplication in the central nervous system are common in the latter group. Among the enteroviruses, however, only Coxsackie A21 acts primarily as a respiratory virus, and its importance is limited mainly to military recruits. Enterovirus 70 causes acute hemorrhagic conjunctivitis, and the virus is present in the conjunctiva and throat (see Chapter 9).

9.2. Gastrointestinal

Hepatitis viruses, enteroviruses, adenoviruses, reoviruses, and rotaviruses multiply within the gut. Many of the same barriers that prevent cell attachment and penetration may exist there as in the respiratory tract, including local IgA antibody. Local, humoral, and cell-mediated immunity follows natural viral infections of the intestinal tract and is the basis for immunity following oral administration of live vaccines such as poliomyelitis and adenoviruses 4 and 7. Unlike the case with respiratory viruses, local multiplication does not produce local symptoms; these occur only after implantation has occurred in secondary sites of multiplication such as the liver for hepatitis virus and the central nervous system for enteroviral infections. Exceptions are the rotavirus and parvovirus (Norwalk agent) infections of children[46] and adults (see Chapter 11 and references 54, 86, and 118).

9.3. Systemic Infections

Systemic infections involve viremia with or without additional spread along other routes. Spread via the bloodstream is the major route by which many viruses locate in secondary habitats, where their principal effects are produced. Some viruses become closely associated with lymphocytes in the bloodstream during the viremic phase and may persist there for years; these include CMV, EBV, human retroviruses, measles, and poxviruses. Some produce a chronic proliferative infection of B lymphocytes (EBV), and some of T lymphocytes (HTLV-1); others cause destruction of T_4 lymphocytes (HIV or HTLV-III/LAV). Some are free in the plasma (arboviruses, enteroviruses, hepatitis vi-

ruses) or circulate as immune complexes. Some have a special affinity for red blood cells (Colorado tick fever and Rift Valley fever viruses). Viremia may be maintained by continual or intermittent seeding from the liver, spleen, bone marrow, and other organs. The persistence of CMV, EBV, human retroviruses (especially HIV), and hepatitis viruses in the blood for years poses a hazard in their transmission via blood or blood products. Most of these occur when the viruses circulate in the presence of antibody. Persistent antigemia may result in other consequences. Immune complexes may form, deposit, fix complement, and cause local tissue injury, especially in small blood vessels as in HBV and periarteritis nodosa; HBV antigemia may also result in hepatocellular carcinoma with or without an intervening cirrhosis. Prospective studies in Taiwan have shown a 223-fold increased risk of hepatocellular cancer in those with antigemia as compared with those without.[12]

9.4. The Exanthem

Our understanding of the pathogenesis of systemic infections associated with a rash such as the pox group, measles, and rubella has been enhanced by the fine studies of Fenner with mousepox.[82] In each such exanthem, there is an incubation period of 10–12 days before symptoms of illness appear. After multiplication of the virus at the site of implantation and in the regional lymph nodes, a primary viremia occurs within the first few days, resulting in seeding of organs such as the liver and spleen. A secondary viremia then follows, with focal involvement of the skin and mucous membranes, the appearance of a rash, and the onset of symptoms. In mousepox, a primary lesion then develops at the site of inoculation. Although the destruction of cells involved in viral multiplication and the release of pyrogens from leukocytes may be responsible for symptoms such as fever, the appearance of antibody at this time suggests that antigen–antibody complexes may play an important role in the pathogenesis of the rash. The viruses of smallpox, herpes simplex, and varicella–zoster are present in the skin vesicles of each of these diseases.

9.5. Infections of the Central Nervous System

In a comprehensive review of the pathogenesis of viral infections of the central nervous system (CNS), Johnson and Mims[115] emphasize that one or more routes of infection may be involved and that the pathways differ with the particular viruses, the host, and the portal of entry. In man, the hematogenous routes to the CNS from the portal of entry and from primary multiplication sites in the gut, respiratory tract, parotid, or lymph nodes are clearly of importance in enteroviral infections, mumps, lymphocytic choriomeningitis, primary herpes simplex infections, HIV infections and fetal infections with rubella virus and CMV. Secondary multiplication sites in the liver, spleen, muscle, or vascular tissue may augment or maintain the viremia; the brown fat has also received attention in this regard for a variety of viruses. Several mechanisms have been suggested as to how viruses enter the brain from the bloodstream. This may be a passive process, or the viruses may actually grow their way through the choroid plexus. Viral multiplication at this site or leakage into the cerebrospinal fluid following growth in the meningeal cells may explain the presence of echovirus and coxsackievirus in the spinal fluid during CNS infections; the presence of viral-specific IgM antibody in the spinal fluid usually indicates active viral multiplication in the CNS.[113,114]

The blood–brain barrier is represented morphologically by the cerebral capillaries, whose endothelial cells lack fenestrations, are joined by tight junctions, and are surrounded by dense basement membranes.[114] This barrier inhibits viral invasion of the CNS and may deter viral clearance. The blood–brain barrier also isolates the CNS from systemic immune responses in the absence of disease, and in normal persons the immunoglobulins present in the cerebrospinal fluid (CSF) are derived from the blood and are dependent on the size of the immunoglobulin molecule: IgG and IgA are present at about 0.2 to 0.4% of the plasma levels, and IgM at a lower level.[114] During an inflammatory process, there is a change in the blood–brain barrier allowing transudation of serum proteins, including immunoglobulins. Once plasma cells are recruited to the CNS, synthesis of immunoglobulins occurs in the CNS, but of limited heterogeneity. T lymphocytes are also recruited that are sensitized to the invading virus and release lymphokines that attract macrophages; these constitute the majority of cells in the inflammatory response.

Neural spread along nerves can occur in rabies, poliomyelitis, and B virus infections of man. In rabies, it appears to be the predominant, if not the sole, method of spread to the CNS, whereas it seems to be relatively unimportant in poliomyelitis. The axons, lymphatics, and tissue spaces between nerve fibers represent three possible conduits for spread along the neural route. Transmission via the tissue spaces plus direct infection and involvement of endoneural cells seem the most likely mechanisms. Spread along the olfactory pathway has also been experimentally demonstrated for poliomyelitis, herpes simplex, and certain arthropod-borne viruses. The role of this route in natural infections is uncertain. As with respiratory viruses, those that infect the CNS have different cell preferences: poliomyelitis has a predilection for anterior horn cells of the spinal cord and the motor cortex of the brain, and arboviruses have a predilection for cells of the encephalon. Herpes simplex appears to have more catholic tastes and multiplies in a wide variety of cell types. As is also true of respiratory cells, the existence of specific cell recep-

tors for individual viruses may play a crucial role in susceptibility.

9.6. Persistent Viral Infections

The pathogenetic mechanisms discussed thus far have dealt with infections in which an acute illness results, usually after a relatively short incubation period (except for rabies), and in which recovery ensues. The virus disappears and is often eliminated from the body. Another pathogenetic mechanism under increasing study is one in which the virus persists for months or years and may result in delayed host responses. Some of these persistent viruses are also capable of evoking an acute response; these include the herpesviruses, rubella virus, the adenoviruses, measles virus, and other paramyxoviruses. Other persistent viruses such as papovaviruses and polyoma viruses rarely produce any acute illness.

In persistent viral infections of the CNS, prolonged synthesis of specific IgG immunoglobulins may be found, as in subacute sclerosing panencephalitis (SSPE) or rubella panencephalitis. Included in this group of potentially persistent viral agents are the herpesviruses, adenoviruses, papovaviruses, paramyxoviruses, rhabdoviruses, retroviruses, coronaviruses, arenaviruses, togaviruses, and picornaviruses, all of which have been shown capable of long-term neural infections.[113]

Still other agents called "slow viruses" produce chronic degenerative disease years after exposure. This group includes kuru and Creutzfeldt–Jakob disease of man, scrapie infection of sheep, transmissible mink encephalopathy (see Chapter 25) and the lentiviruses, of which HIV appears to be a member.

Six factors that favor persistence of certain viruses have been summarized by Mims[151]: (1) persistent viruses tend to have low or no pathogenicity for the cells they infect, in contrast to viruses with severe, destructive effects, which induce acute disease terminated by death or by recovery and the elimination of the virus; (2) there may be an ineffective antibody response possibly because of tolerance, autoimmunosuppression, production of nonneutralizing or blocking antibodies, not enough antigen on the surface of the infected (target) cell to induce adequate antibody formation, or spread of the virus directly from cell to cell where antibody does not reach it; (3) there may be an ineffective cell-mediated immune response for reasons similar to those involved in the poor antibody response [tolerance, autoimmunosuppression, blocking antibodies, too little antigen expressed on surface to infected cell, failure of immune cells to reach infected (target) cells]; (4) there may be a defective interferon response, such as in lymphocytic choriomeningitis in mice; other viruses may be relatively insensitive to interferon action even though it may be produced; (5) certain persistent viral infections induce neither an immune nor an interferon response; these include the "slow virus" infections such as kuru and Creutzfeldt–Jakob disease; (6) lymphocytes and macrophages are often infected in persistent viral infections, such as with adenoviruses, EBV, CMV, and measles virus, thus altering the host's immune response. Interferon produced by infected macrophages may have no protective effect on other macrophages, although there is normal activity on normal cell types; certain virus–antibody complexes still remain infectious after phagocytosis by macrophages; infected macrophages may be less active in releasing the same virus from the blood, thus favoring persistent viremia.

Such persistent and latent viral infections may reactivate, producing the acute disease again, or may result in a chronic viral infection manifested by immune complex disease, degenerative diseases of the CNS, or certain malignancies. These infections will acquire greater visibility and importance as immunosuppressive drugs are used more widely in medical therapy and in organ-transplant recipients. They now include the consequences of AIDS in which acute, chronic, and malignant manifestations may appear as a result of the reactivation of many types of microorganisms, including viruses, some of which are nonpathogenic in the normal host. Certain genetic disorders involving the immune system can also result in persistent and/or reactivated viral infections or in aberrant responses to them such as the X-linked lymphoproliferative syndrome.[179]

10. Incubation Period

The period from the time of exposure to the appearance of the first symptoms is called the *incubation period.* Viruses that do not require distant spread but are able to produce disease through multiplication at the site of implantation, such as the respiratory tract, have short incubation periods of the order of 2–5 days. Those that require hematogenous spread and involvement of distant target organs such as the skin or CNS have incubation periods of 2–3 weeks.

In AIDS the incubation period may be 4 to 7 years or more and is dependent on the time it takes for destruction of T_4 lymphocytes by the virus to permit opportunistic infections to become pathogenic or for latent agents to reactivate and produce clinical disease. Recently, an earlier, primary repsonse to the virus has been recognized occurring 3–4 weeks after infection and characterized by fever, malaise, lymphadenopathy, rash, headache, and stiff neck.[96] In kuru, the incubation period after exposure by ingestion of infected brain or other tissues or by absorption via abraded skin at a cannibalistic feast ranges up to 27 years or more.[91]

Viruses such as rabies, dependent on spread along nerves, have very long and variable incubation periods ranging from 8 days to a year or more. The variation in incubation

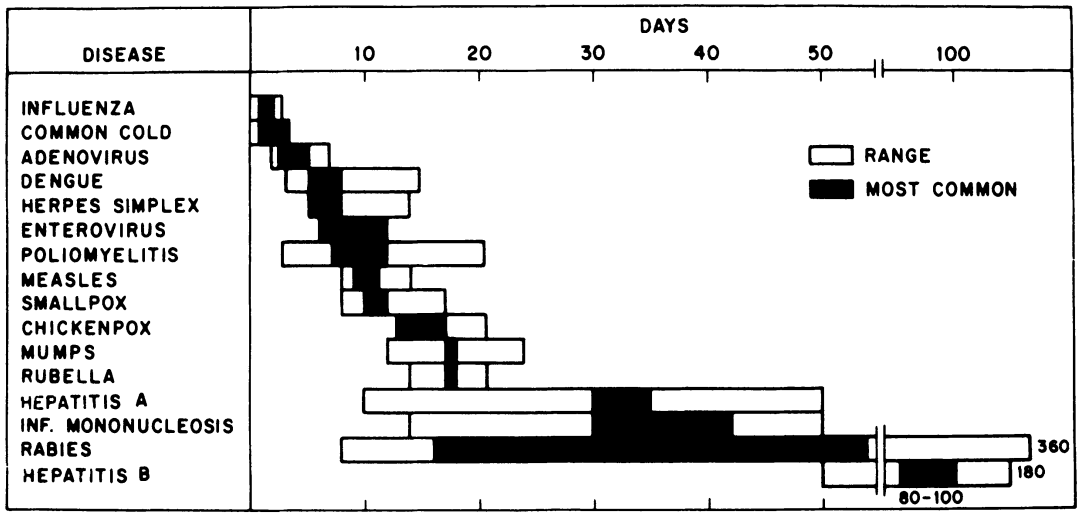

Figure 1. Incubation periods in viral diseases. Based mostly on data from Benenson. [16]

periods in different diseases is indicated in Fig. 1. In some diseases, early symptoms or even a rash may accompany the period of initial invasion or viremia. This has been seen in poliomyelitis, dengue, hepatitis, and infectious mononucleosis. In such instances, the apparent incubation period to the appearance of these early features is much shorter than the usually accepted period; more often, this early phase is not clinically recognized or occurs before the patient visits the physician.

Knowledge of the incubation period has many practical uses. Epidemiologically, it helps define the period of infectiousness: a patient is not usually infectious until close to the time of the appearance of clinical symptoms. In epidemics, knowledge of the mean, minimum, and maximum incubation periods can be used to identify the probable time of exposure to the index case or other source of infection. The duration of infectivity depends on the persistence of the virus and its exit into the environment. Clinically, the duration of the incubation period helps to identify the likelihood of viral exanthem after a known exposure or to differentiate hepatitis A from hepatitis B infections. Prophylactically, it determines the feasibility of prevention of the clinical illness by immune serum as in hepatitis A, varicella–zoster infections, rubella, and rabies, as well as the potential success of rabies vaccination.

In addition to the viruses that produce acute infections, there are delayed effects of certain common viruses in which the "incubation period" may be several years. An example is the relationship of measles virus to subacute sclerosing panencephalitis, in which infection in infancy may be associated with involvement of the CNS some 5–10 years later.[27]

Certain papovaviruses cause widespread inapparent infections in childhood. Rarely, reactivation occurs later in life in the form of progressive multifocal leukoencephalopathy. This is seen in patients with Hodgkin disease in association with depression of cell-mediated immunity (see Chapter 25). The term "incubation period" may be inappropriate in this setting because viral reactivation appears to be involved and not a period of primary multiplication of the virus.

The concept of an incubation period has also been applied to putative oncogenic viruses and to chemical carcinogens in relation to the subsequent development of cancer. In various neoplastic diseases, the estimated incubation period from a defined point of exposure to a suspected carcinogen to the development of disease has ranged from 0.27 year for the development of pancytopenia after chloramphenicol to 36 years for the appearance of lung cancer after exposure to asbestos.[8]

11. The Immune Response

The immunologic response of the host to a virus infection plays a role not only in the development of specific resistance but also in the pathogenesis of the signs and symptoms of the disease itself.[164,165] Specific immunity to viral disease is based on humoral antibody, local antibody, and cell-mediated immunity. It is evoked either by natural infection or by immunization with live or killed antigens. The immune responses are dependent on B-type lymphocytes derived from the bone marrow and on T lymphocytes derived

from the thymus in cooperation with macrophages. The field of immunology is a complex and very rapidly expanding one. The purpose of this brief and superficial account is only to encourage the reader to seek other texts to understand more fully the important role of this subject in infectious disease epidemiology and to enhance an understanding of the concepts of pathogenesis and immunity on which we base much of our discipline.[3,4,47,49,98,112,150,151,161–168]

11.1. Humoral Immunity

Humoral immunity is dependent on antibody found in the blood and other body fluids. Of the five classes of immunoglobulins, IgG, IgM, and IgA are important in this type of potection.

Virus-specific IgG is the major circulating type and usually persists for life, and the molecules are capable of passing the placenta of pregnant women, conferring temporary immunity on the newborn. The molecule is composed of two heavy and two light polypeptide chains: the antigen-reactive ends of the light and heavy chains are responsible for its specificity. The Fc fragment has no antigen-reactive sites but binds complement, binds to receptors on the surface of polymorphs and macrophages, and mediates attachment of antibody-coated microorganisms to the phagocyte. Some IgG is also present in other body fluids such as lymph, peritoneal, synovial, and cerebrospinal fluids.

Immunoglobulins of the IgM class appear in response to the primary infection, are of relatively short duration, over 3–4 months, and are commonly taken as a marker of a recent infection. They may reappear in lower titer in some reactivated viral infections, especially in the herpes group, and may sometimes persist over longer periods as in congenital infections or in the brain in measles infections early in life that may lead to subacute sclerosing panencephalitis. They remain confined to the vascular system except where they are produced locally as in infections of the CNS. Molecule for molecule, they have five times the number of antigen-reactive sites as IgG; the IgM molecule also has five times the number of Fc sites and therefore five times greater capacity to activate complement.

Immunoglobulin M antibody appears first in response to infection and thus may play a determining role on the course of the infection; it is then followed by IgG antibody. The presence of these antibodies in the blood constitutes a major deterrent to the spread of viruses to distant sites. They constitute the major basis for vaccines that induce humoral immunity and prevent the development of clinical illnesses dependent on viremic spread, such as poliomyelitis, hepatitis, and the viral exanthems. Infections characterized by viremia also produce the most marked and longest-lasting humoral antibody response.

Three types of cells are involved in antibody formation. The B cell, derived from the bone marrow, is the producer or effector cell that synthesizes and secretes antibody. For most antigens, lymphocytes, which are processed through the thymus gland, are required, primarily the helper (h) or T_4 lymphocyte. The third cell required is a nonlymphocyte cell, the macrophage cell. This is involved in the processing and presentation of the antigen to the lymphocyte, an interaction dependent for recognition on the presence of the major histocompatibility complex (MHC). The absence of any of these three cells will impair or abolish the antibody response.[97]

Passive transfer of convalescent serum or immune globulins may also produce protection against those infections that are dependent on a viremia to reach target organs for their clinical expression. The amount of protection reflects the antibody level of the donors of the blood from which the serum or globulin is derived, and passive antibody does not protect against multiplication of the virus at the site of initial implantation in the respiratory or gastrointestinal tract.

11.2. Local Immunity (Secretory IgA System)

Local antibody production is mediated through virus-specific immunoglobulins of the IgA class. These secretory IgA molecules are the principal ones on mucosal surfaces and in milk (colostrum). They are critical in resistance to infection in the respiratory, intestinal, and urinogenital tracts. Their production follows natural infection or administration of live vaccines by the natural portal of entry. Less effective production occurs with live vaccines given parenterally, and usually poor responses occur with killed vaccines given parenterally; similarly, administration of passive antibody or γ-globulin does not provoke local immunity. This leaves these unprotected mucosal surfaces susceptible to primary infections with viral agents entering these portals. Although the individual may be protected against clinical illness by humoral antibody, epidemiologically the multiplication and excretion of the organism provides a continued method of spread in the community. Thus, protection against clinical disease but not against infection for the individual or his or her contacts may result from vaccines that fail to induce a satisfactory secretory IgA antibody response. In the submucosal tissues, antigens combine with specific antibody, form immune complexes, enter the blood, and are then filtered out and excreted in bile. The IgA molecule lacks the secretory piece and, after responding to local antigens, enters the blood via lymphatics to give increased serum IgA levels. The IgA-producing cells (B lymphoblasts) may also be carried via lymph and vascular routes to areas other than the local site such as the salivary glands, lung, mammary glands, and intestine or even to other sites of the same organ, i.e.,

other parts of the intestinal tract. There, they may be active in preventing local infection in these new locations.

11.3. Cell-Mediated Immunity

Delayed hypersensitivity is a classic manifestation of cell-mediated immunity as exemplified by positive skin-test reactions. In viral infections, lymphocytes and the lymphokines they produce attract macrophages and, with their products, participate in the destruction of antigen and antigen-infected cells. The T lymphocyte is of key importance in the recognition and management of viral and other intracellular infections.[2,3,97,98,151,163–166] When T cells are absent, depleted, or functionally impaired, severe and widespread viral, fungal, or intracellular bacterial infections may develop, latent agents may reactivate, and organisms usually considered nonpathogenic may result in illness. This situation is exemplified by the lysis of T_4 (helper) lymphocytes by HIV infection leading to AIDS, AIDS-related complex, chronic neurological disease, and various malignancies. The existence of the T-cell receptor has now been established, and in conjunction with MHC recognition, it provides the basis for T-cell activation when presented with a specific antigen by macrophages. Antiidiotype antibody against specific T-cell receptors is being pursued as a means of preventing antigen attachment and in this way providing immunity without use of the antigen itself.

There are different subclasses of T lymphocytes that play different, and sometimes opposite, roles in the immune response. These include suppressor T cells, helper T cells, and cytotoxic (killer) T lymphocytes. These functions are usually highly antigen specific, but some killer T cells have nonspecific cytotoxic activity. Macrophages are involved in the immune response and are activated by lymphokines, by immune complexes, or by the third component of complement; these activated cells act nonspecifically to limit viral multiplication, effect complement cleavage, and carry out phagocytosis. In addition to monophages, there is another cell capable of killing, the so-called K cell, which is lymphoidlike but of uncertain lineage. It has a receptor for the Fc portion of IgG and can kill target cells covered by antibody. Increasing attention is being focused on the T suppressor or immunoregulatory cells. Effector responses of both B and T cells are controlled by these regulator cells; the appropriateness of their activities is under genetic control, probably from loci on the sixth chromosome. Subtle alterations in their effectiveness may lead to disease states, possibly to malignancy.

Although the prevention of spread of viruses through extracellular fluids and the blood seems to be largely dependent on neutralization by humoral antibody, control of viral spread from cell to cell is probably dependent on cellular immunity. The latter form of contiguous infection might be interrupted by destroying infected cells, by severing connections between infected and uninfected cells so that the virus cannot be transferred to uninfected cells without being exposed extracellularly to neutralizing antibody, or by destroying contiguous uninfected cells so that virus must proceed extracellularly to reach target cells for further multiplication.[162,164] An important element in the destruction of infected cells is the induction of virus-induced antigens on the cell surface, which makes them appear foreign to other host cells; they are then destroyed by T-type lymphocytes as in graft-versus-host rejection.

Knowledge of the mechanism of T-cell activity and cell killing is rapidly being attained. Following infection by a virus, there is an early nonspecific defense system, then one dependent on virus-specific antibody and lymphocyte responses. In the early response a group of cells called "natural killer cells" or NK cells play a key role[29]. These are regarded as large granular lymphocytes and represent some 5–15% of peripheral blood lymphocytes in humans. They possess spontaneous cytotoxic activity and can lyse virus-infected as well as neoplastic and even normal cells without prior exposure to specific antigens. The NK cells also express surface receptors for the Fc portion of IgG and act as effector cells in antibody-dependent cell-mediated cytotoxicity (ADCC) but require the presence of specific antibody, which does not appear until later.

The specific effector arm of cell-mediated immunity is dependent on a subset of lymphocytes requiring 7 to 10 days for peak activity and involving helper/delayed-type hypersensitivity or suppressor/cytotoxic phenotype. On activation of lymphocytes by a specific antigen, they proliferate and release a group of soluble substances or mediators. These are termed lymphokines and include interleukin 2 (IL-2) or T-cell growth factor, interferon, macrophage inhibitory factor, macrophage-activating factor, colony-stimulating factor, and lymphotoxins. A single lymphokine may have more than one activity, and any single activity may be triggered by more than one lymphokine. Stimulation of lymphotoxins is antigen specific, but its effect is spread over several types of lymphocytes, fibroblasts, and tumor cells.

Killer cells apparently release proteins, called "perforins," that produce holes or pores in target membranes.[143] Lymphotoxins may enter through these holes and break up target cell DNA.[184] Another protein called tumor necrosis factor is also involved in killing of target cells; it is produced by macrophages and is structurally related to the lymphotoxins.

The specific events in the immune response to a viral infection may best be illustrated by a specific example, that of herpes simplex, one of the best studied viral agents.[128] When the virus breaches the barrier of the normal skin or

mucosal membrane, a rapid inflammatory response occurs including both polymorphonuclear cells (PML) and mononuclear cells; interferon is generated by local fibroblasts, as are other as yet unidentified antiviral substances. This early phase is termed containment. Free virus is phagocytized by macrophages, which may allow early steps in viral replication but prevent final viral assembly and production of mature particles. A second challenge is control of the virus-infected cell. Shortly after infection of the cell, it acquires new viral antigens on the surface and pours out virus or transfers it by intracellular bridges from cell to cell, thus avoiding marauding phagocytes and specific humoral components that are soon to appear. In this early phase the NK cell destroys HSV-infected cells or decreases viral production without cell lysis. Interferon enhances these effector mechanisms while appearing to protect normal cells against NK cell lysis.

The second phase is that of specific effector control or eradication, which is mediated through two major systems that become active during the middle of the first week of infection and last until replicating virus is eradicated. These are the T-cell-mediated effector functions and the B-cell- or antibody-mediated effector functions. Protection by T-cell activity is carried out by a variety of effector mechanisms including direct T-cell cytotoxicity restricted by type 1 transplantation antigen (H-2 or HLA), delayed-type sensitivity, and effector lymphokines such as interferon, IL-2, and other antiviral agents such as lymphotoxin. Humoral defense mechanisms are also activated but are dependent on helper T cells. Antibody production appears to be extremely heterogeneous in regard to both the array of recognized epitopes and the functional type of antibody generated.[128] The lack of production of a specific set of idiotypes may determine the severity of illness or the frequency of recurrences.

Free virus can be neutralized by antibody alone, but for virus-infected cells, ADCC is critical in conjunction with complement and leukocytes. Herpes simplex virus also programs the appearance of an Fc receptor on the surface of the infected cell that is able to bind immunoglobulin, but the precise role of this receptor is presently poorly understood.

In the normal human the viral infection is contained smoothly, leaving the host with a low-level but life-long latent infection. In the neonate, however, a much more severe and sometimes fatal infection occurs because of a variety of defects in the proper functioning of the immune system. These include the permissiveness of unstimulated lymphocytes for HSV replication, the paucity of the macrophage barrier, and a profound defect in NK cell activity. Humoral defects are also present, including a poor and often delayed antibody response combined with a helper T-cell defect and probably low levels of antigen-specific T cells themselves. Knowledge of the operation of the immune system and of its mediators may permit the eventual reconstitution of defective elements through bulk cultures and clones, as presently accomplished in mice.

11.4. Immune Responses in the Pathogenesis of Viral Diseases

Viral infections may produce the symptoms of disease through a variety of mechanisms, some of which are immunologic in nature.[49,163–166] Antibody produced by the virus may circulate until it reaches the virus and in combining with it initiate an attack on the tissue to which the virus is attached. Viruses may circulate in the blood, forming circulating immune complexes with the antibody they have induced. The consequences of this depend on the antigen–antibody balance and the size of the complex formed.[49] With large antigen excess, the complexes are small, are excreted readily, and do not activate complement. With antibody excess, large complexes are formed that are phagocytized and removed. The pathogenic complexes are those in balance or with slight antigen excess that combine with complement and deposit in blood vessels, especially in the glomeruli of the kidney. Together with polymorphonuclear cells, they may evoke an inflammatory response and tissue injury. Immune complex nephritis is the best-studied example of this. A third mechanism of injury, referred to previously, is the induction by viruses of new antigens on the surface of the cell. These neoantigens are regarded as foreign by host cells and may evoke antibody formation and a cell-mediated response that results in host cell injury or in immune complex formation. If the virus-infected cell is a lymphocyte, as in the EBV infection causing infectious mononucleosis, then the neoantigen induced on the B cell may result in a mixed-lymphocyte response with T-cell transformation and proliferation.[11,141,206] In this situation, the atypical lymphocytosis characteristic of the disease may result both from virus-transformed B cells and from T cells entering blast formation as an immune response to altered B cells. A fourth mechanism of immune viral injury might occur when the virus or the virus-induced antigen shares a common component with normal tissue and an autoimmune response results.

Finally, a fifth mechanism is that viral infection of a cell can induce functional changes without causing cell pathology.[166] Alterations in the production, characteristics, or release of cellular products may occur without resulting in death or detectable injury of the infected cell, yet result in disease.

Our knowledge of cell-mediated immunity and cell-mediated tissue injury is incomplete. An increasing understanding of the mechanisms involved, of the relationship of cellular to humoral immunity, and of the consequences of depressed immunity may explain why certain viruses persist

and how such persistence may relate to cancer, immune complex diseases, and chronic infections of the central nervous system.

12. Patterns of Host Response

The host responses to viral infections vary along a biological gradient in terms of both the severity and the nature of the clinical syndrome produced.

Although the emphasis on the biological gradient presented here is on the entire host, it is clear that different qualitative and quantitative responses may occur at the cellular level. Biochemical changes in the molecular composition of the virus, even a change in a single nucleotide, may alter its effect on susceptible cells, and genetic alterations in the host cell affecting the presence or absence of specific receptors for viral attachment and entry and probably the internal assembly and release of the viral particle may affect the nature and gradient of the cellular consequences of viral infection.

12.1. The Biological Gradient

The host response to a virus may range from a completely inapparent infection without any clinical signs or symptoms to one of great clinical severity, even death. The ratio of these inapparent (or subclinical) to apparent (or clinical) responses varies from one virus to another; representative examples are shown in Table 4. At one end of the spectrum are certain infections that are almost completely asymptomatic or unrecognizable in their pattern until some special event provokes a clinical response. The response may appear long after the initial infection and arise from viral persistence or reactivation or both. The BK and JC strains of papovavirus fall into this category: no known clinical disease has been associated with the initial infection, which is a common one in Wisconsin school children as reflected by a 70% prevalence rate of antibody to the virus.[171,190] However, this virus becomes reactivated in persons with Hodgkin's disease and others in whom there is a depression of cell-mediated immunity, leading to the development of a fatal disease of the central nervous system known as progressive multifocal leukoencephalopathy (PML).[172] The virus can be isolated from the brains of such persons, and high antibody titers may be present if the person survives long enough.[172]

The subclinical : clinical ratio for the acquired immunodeficiency syndrome (AIDS), AIDS-related complex, and other manifestations is not included in Table 4 because it is not clearly enough defined at this time. It is dependent in part on the diseases that represent a primary result of infection with HIV, such as an early febrile disease with rash and stiff neck and later direct neurological consequences of primary infection, and mainly on the presence of opportunistic and latent agents present in the infected individual. It also varies with the presence of other coexisting infections, the risk group involved, the geographic area, and the age at which infection occurs. This issue of the clinical expression among persons with HIV antibody is of great public-health importance because, as of 1988, it is estimated that over a million and a half persons have been infected with the virus in the United States alone, and the long-term mortality approaches 100%. As of this writing, there is no vaccine and no effective antiviral agent or other compound to prevent the development of the disease in infected persons or cure them once clinical symptoms have appeared, so the situation is one of

Table 4. Subclinical/Clinical Ratio in Viral Infections (Inapparent/Apparent Ratio)

Virus	Clinical feature	Age at infection	Estimated subclinical/clinical ratio	Percentage of infection with clinical features
Poliomyelitis	Paralysis	Child	±1000 : 1	0.1–1
Epstein–Barr	Heterophil-positive infectious mononucleosis	1–5	>100 : 1	1
		6–15	10–100 : 1	1–10
		16–25	2–3 : 1	50–75
Hepatitis A	Jaundice	<5	20 : 1	5
		5–9	11 : 1	10
		10–15	7 : 1	14
		Adult	2–3 : 1	50–75
Rubella	Rash	5–20	2 : 1	50
Influenza	Fever, cough	Young adult	1.5 : 1	60
Measles	Rash, fever	5–20	1 : 99	99+
Rabies	CNS symptoms	Any age	0 : 100	100

great magnitude. It is one requiring vigorous health education to protect the uninfected and to identify the infected to prevent further spread.

Current estimates of the development of clinical illnesses per year in HIV-infected persons have been 5.9% in high-risk homosexuals, 3.5% in low-risk groups, 2.5% in high-risk I.V. drug abusers, and 2–4% in hemophiliacs.[23] Prospective studies of HIV-infected and high-risk homosexual males over a 3-year period have revealed an incidence of AIDS of 34.2% in New York City, 17.0% in Washington, DC, and 12.5% in Denmark; in high-risk I.V. drug users in Queens, NY, the rate has been 12.5%, and in hemophiliacs in Hershey, PA, it has been 12.8%. Over 7 years, 50–80% may show some clinical manifestation of HIV infection.

In Africa, high antibody prevalence has been found in many areas, but with great geographic variation and with an equal prevalence in males and females. The number of reported clinical cases of AIDS has been very low. Whether this reflects lack of clinical and laboratory facilities to diagnose the disease, failure to report it, political issues, a low rate of clinical illness when infection occurs early in life, or an attenuated strain of the virus is currently unknown. There is hope that an attenuated strain, or isolates from simian sources, might be the basis for a vaccine, even though great antigenic variation in HIV isolates have been identified. However, as of this writing in mid-1988, the prospect does not look very encouraging because of antigenic variation in the human strains thus far isolated.

Chemoprophylactic and chemotherapeutic drugs that act at different sites in the replicative cycle of the virus or in its attachment to the cell are also under development and clinical testing with the hope that they might be used to block the subsequent development of AIDS and related complexes among the large number already infected (see Chapter 21 on retroviruses for further details). Current efforts must therefore focus on preventing spread among the already infected and preventing the uninfected from entering high-risk activities.

A second group of viral infections are those that are predominantly mild or asymptomatic when exposure and infection occur in early childhood but that frequently result in symptomatic and sometimes severe clinical disease when infection is delayed until late childhood and young adult life. Examples of this are viral hepatitis, poliomyelitis, and EBV infections.

At the other end of the spectrum are infections caused by measles, rabies, and Lassa fever viruses, in which clinically recognized illness usually accompanies the infection. Indeed, in rabies infection of man, death is almost inevitable after characteristic symptoms develop.

This biological gradient of host response is often pictured as an iceberg in which clinically apparent illness—i.e., above the water line—represents only a small proportion of the response pattern and the larger amount represents unrecognized and inapparent infections; a similar analogy may exist at the cellular level. Figure 2 portrays these concepts. The cellular responses shown might better be considered as

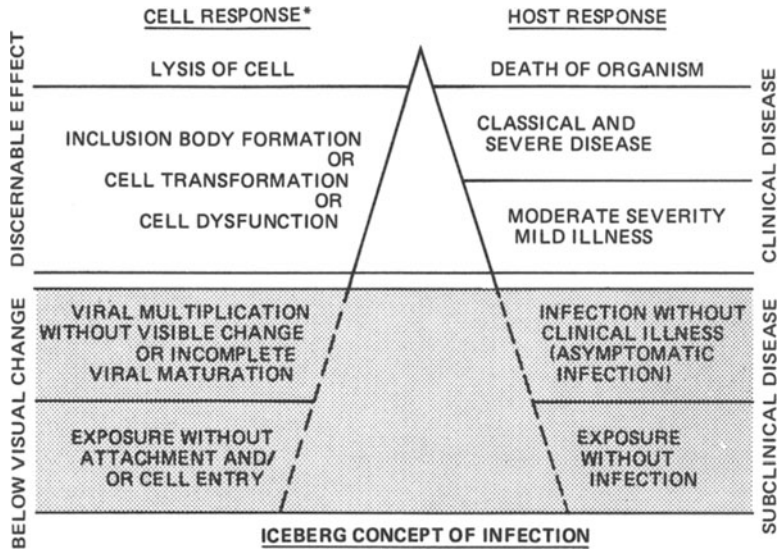

Figure 2. "Iceberg" concept of infectious diseases at level of the cell and at level of the host. Within any cell population, varying patterns of cell response also occur. *, Hypothetical.

differences in the nature rather than the severity of the response.

12.2. Clinical Syndromes

The nature and severity of the host reponse vary widely in viral infections, even with the same virus. These various clinical responses may reflect variations in the strain of virus, even minor biochemical differences, different organ tropisms of the virus, different portals of entry, different ages at the time of infection, variations in the immune response, and differences in the genetic control of the characteristics of the agent and of the immune response of the host to it. The clinician faced with the diagnosis and management of a patient presenting with clinical syndromes involving various organ systems, or with a rash, may have great difficulty in making an etiologic diagnosis based on clinical features alone, even in distinguishing between viral and bacterial infections. This is because these target organs have only a limited number of ways to respond to infection, and any one of several viruses or other causative agents may trigger the same general response pattern. These causes will also vary with age, season, year, and geographic setting. The results of specific viral isolations and of serological tests may come too late to be useful during the acute illness, although advances in rapid, direct identification of many viral agents and demonstration of virus-specific IgM antibody are rapidly changing this situation. Such tests are often available in special laboratories.

Early knowledge of the viral etiology of a syndrome may avoid misuse of bacterial antibiotic therapy and for some viral infections may allow selection of an appropriate antiviral compound. Prevention of infection in exposed and susceptible contacts may also be possible. Often, however, the physician must rely on epidemiologic and clinical features and simple laboratory tests in making a tentative etiologic diagnosis. This diagnostic reasoning is based on the known frequency of a given causative agent in that year, season, age group, or special setting and its epidemic behavior. The following sections present some of the etiologic agents involved in common clinical syndromes, sometimes in the form of a "pie diagram," but their limitations should be kept in mind.

12.2.1. Common Respiratory Syndromes. A great many viruses and viral groups can evoke respiratory symptoms and diseases, as can bacteria, rickettsiae, and certain fungi. The viral causes vary from season to season, from year to year, from place to place, between and within countries, and especially from one age group to another.[125] The etiology differs between infancy and childhood. Few studies have evaluated both viral and bacterial causes in the same population group at the same time and setting. Although studies of viral etiology have included minor and major illnesses within the family, community, and hospital, those of bacterial origin have focused mainly on the more severe and hospitalized cases. Lung aspirates have given different results from those of sputum or of oral/pharyngeal washings.[124] Thus, the generalizations made in this discussion must be accepted with caution. In general, however, the great majority of total respiratory illnesses in infants, children, and young adults are caused by viruses with the exception of *M. pneumoniae* in older children and young adults. In the more severe and hospitalized cases, and in those over 50 years of age, bacterial infections play the predominant role.

A number of investigators have tried to sort out the varied etiology of clinical syndromes of acute respiratory diseases in different age groups and population settings.[16,45,56,60,95,140,155,195]

In infants under 2 years old, respiratory syncytial virus (RSV) is the most important respiratory pathogen, producing bronchitis and bronchiolitis as well as pneumonia, croup, otitis media, and febrile upper respiratory disease. *Chlamydia trachomatis* has been recognized in some studies as an important cause of pneumonia in the first 6 months of life.[103] Parainfluenza virus type 3 is second to RSV as a cause of pneumonia and bronchiolitis in infants less than 6 months of age. Both viruses can reinfect and cause upper respiratory illnesses in older children and adults. Parainfluenza type 1 is the most important cause of croup (laryngotracheobronchitis) in children; type 2 resembles type 1 in clinical manifestations but less commonly causes serious illness. Parainfluenza 4 infections are encountered infrequently. Influenza and adenoviruses also cause acute respiratory diseases in children and young adults.

"Etiologic pies" for four common respiratory syndromes of young adults are depicted in Fig. 3. A fair percentage of the causes remain unidentified. In unimmunized military recruits, adenoviruses types 4, 7, and 21 are important causes of pneumonia and upper respiratory infections. Type-specific vaccines have been effective by oral administration in preventing adenovirus infections in these high-risk populations. *Mycoplasma pneumoniae* is probably the most important cause of acute lower respiratory infections in older children and young adults. Influenza is of importance in all age groups, but the mortality is most associated with infections in infancy and in the aged: this can be caused by primary viral pneumonia, concomitant bacterial infection, or secondary bacterial infection. The predominance of viral infections in infancy and children explains the failure of antibiotic therapy for most respiratory diseases in these age groups. Broad-spectrum antibiotics such as erythromycin are effective in the more serious *M. pneumoniae* illnesses, and penicillin is effective in streptococccal sore throat. Newer antiviral compounds are showing much clinical promise, such as ribavirin in severe RSV infections and amantadine in the prophylaxis of influenza A infections in contained population groups such as nursing homes.

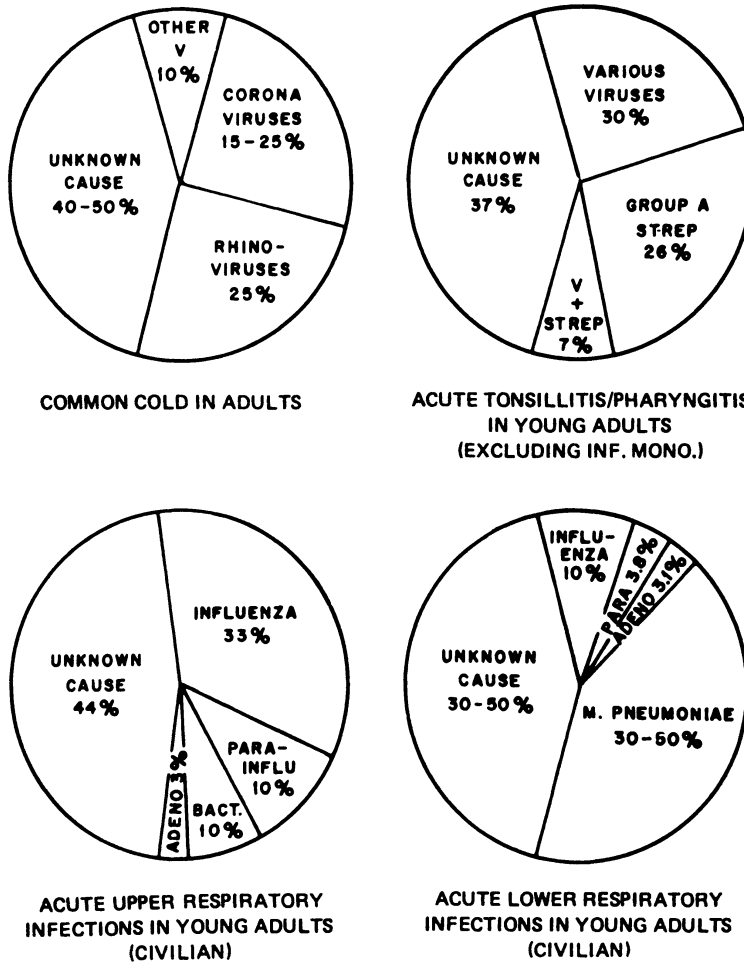

Figure 3. The causes of acute respiratory syndromes in young adults.

12.2.2. Common Infections of the Central Nervous System. Mulitiple agents are also involved in the causation of clinical syndromes of the central nervous system (CNS) as manifested by encephalitis and aseptic meningitis.[113] In 1983 a total of 1795 cases of encephalitis were reported to the Centers for Disease Control (CDC), which represented a 19.7% increase over the 1500 reported in 1982.[33] The increase resided entirely in cases of indeterminate etiology; in 1983 this category accounted for 78.0% of the reported cases. Among the cases of known cause, herpes simplex led the list (Fig. 4) with about 170 cases; this was in part because of the vigor with which this etiology was sought because of its high mortality and because it is the only treatable form of viral encephalitis that responds to therapy. However, the diagnosis requires a brain biopsy, so only more severe and hospitalized cases are likely to be identified as having herpes virus. Enteroviruses accounted for about 50 cases, and the arboviruses for about 70 cases, over half of which were caused by California virus.[198] The last major arbovirus outbreak occurred in 1976 and was caused by St. Louis encephalitis.

The syndrome of aseptic meningitis showed no major change in incidence or etiologic pattern over a period of 5 years, 1979–1983. Yearly fluctuations reflected the activity of the enteroviruses and usually occurred in August or September. In 1983, 12,696 cases of aseptic meningitis were reported to the CDC. No etiologic breakdown was given for these cases, but past experience suggests that about 80% were of unknown or indeterminate cause, some 15% were probably caused by enteroviruses, and a small percent by

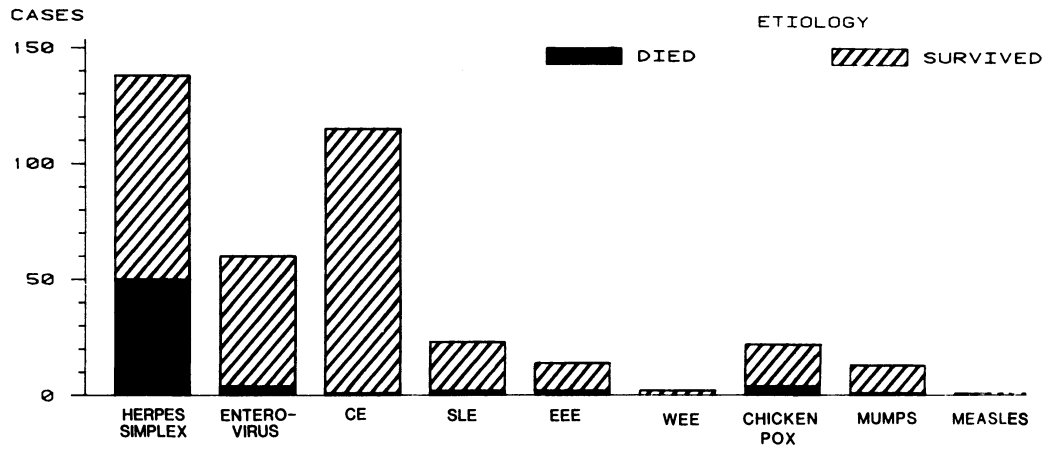

Figure 4. Number of cases and deaths of encephalitis from known causes in the United States in 1983. (CE) California encephalitis; (SLE) St. Louis encephalitis; (EE) eastern equine encephalitis; (WEE) western equine encephalitis. Reprinted from ref. 33.

because of the need in most cases for isolating the virus from the stool, identifying it, and then testing for an antibody rise to that specific agent. In the 1950s and 1960s, certain state laboratories carried out these procedures more intensively, and an etiologic agent was identified in about 65% of the cases.[136] Less intensive investigation in the 1970s resulted in identifying only about 20%. New causes of arborvirus encephalitis such as Snowshoe hare and Jamestown Canyon viruses in North America are being recognized so that a continued monitoring of the etiologic pattern is desirable. Measles encephaliltis is disappearing in the United States because of intensive measles vaccination programs. In tropical countries arboviruses are more common, including a resurgence of yellow fever, and cause many infections involving the CNS.

12.2.3. Common Exanthems. Acute viral syndromes involving the skin are represented by the exanthems of childhood (measles, rubella, varicella, and erythema infectiosa or fifth disease), by various strains of Coxsackie and echoviruses, by certain adenoviruses (such as type 7), occasionally during EBV mononucleosis (often brought on by a reaction to ampicillin), and by the presumed viral cause of roseola infantum (exanthem subitum). A listing is given in Table 5. The newest additions are human parvovirus B19, the cause of erythema infectiosum (fifth disease) as based on demonstration of virus-specific IgM[5] and exanthem subitum associated with human herpesvirus, type 6 (HHV6). The cause of Kawasaki disease, a mucocutaneous lymph-node syndrome, remains unknown, although EBV and a retrovirus have been implicated.[117,123] First recognized in Japan, two outbreaks[13] and many cases are now being seen in the United States and around the world. An unusual feature of the disease is cardiac involvement, with aneurysms of the coronary artery in 17–31% of the cases; the overall case mortality is 0.5–2.8%.

12.2.4. Viral Hepatitis. At least five types of viral hepatitis are currently recognized.[16,130] These include the classical hepatitis A (HAV) or infectious hepatitis and hepatitis B (HBV) or serum hepatitis, delta agent, and at least three non-A, non-B viruses. Diagnostic tests are available for only HAV and HBV and in some special laboratories for delta hepatitis. Reporting to the CDC is based on HAV, HBV, non-A, non-B by exclusion (reported since 1982), and

Table 5. Viral Causes of Common Exanthems

Type of rash	Examples
Macular/papular	Exanthem subitum due to HHV6
	Measles and measles vaccine
	Rubella
	Echo 4, 9, 16
	Coxsackie A9, 16, B5
	Adeno
	Erythema infectiosa caused by papovavirus, type B19
Vesicular	Varicella
	Smallpox
	Eczema herpaticum[a]
	Eczema vaccinatum
	Herpes zoster
	Coxsackie A16
Petechial or purpuric	Coxsackie A9
	Echo 9

[a]A fatal form may occur.[178]

hepatitis, type not specified. Delta virus is an RNA virus present inside the HBV virus and dependent on it for its multiplication. It was first described in Italy but is now recognized in several countries such as Sweden and Kuwait, where most HBV infections are of the delta type and associated with a severe form of the disease. It has also been recognized in Africa and South America and in special risk groups such as hemophiliacs, drug addicts, and male homosexuals. Outbreaks have occurred in Venezuela, an Indian community, and in I.V. drug users in Worcester, Massachusetts. Hepatitis, non-A, non-B, A-type is transmitted by oral–fecal routes, and water-borne outbreaks have occurred in India, Burma, USSR, and other parts of the world. Hepatitis, non-A, non-B, B-type has been found worldwide. In the United States, it accounts for some 90% of posttransfusion hepatitis as well as 15–40% of sporadic community-acquired hepatitis; chronic infections are common and may progress to cirrhosis. There may be two or more types, and a suspect retrovirus is under study (see Chapter 13 for more details). Hepatitis may also occur with EBV and CMV but is not reportable as such.

In 1983 there were 56,499 cases of viral hepatitis reported in the United States, a rate of 24.1 cases/100,000 population. Of these, 38.1% were reported as HAV (9.2/100,000), 61.4% as HBV (10.4/100,000), 6.1% as non-A, non-B (1.7/100,000), and 12.7% as hepatitis, type unspecified (3.1/100,000).

12.2.5. Viral Gastroenteritis. Rapid advances in our knowledge of the causes of acute viral gastroenteritis have occurred over the past few years, and these are presented in Chapter 11. The importance of rotaviruses (formerly termed duoviruses and reolike viruses) as the most important cause of acute gastroenteritis in infants and children under 2 years of age worldwide has now been firmly established through application of a variety of methods to identify the virus in the stool, including immune electron microscopy, the enzyme-linked immunosorbent assay (ELISA), and serological techniques.[18,19,22,26,86,129,148,209] One of the early and seminal studies was that of 378 children with acute gastroenteritis in Melbourne, Australia[46] in which the causes are shown in Fig. 5; rotaviruses (then called duoviruses) were found in the stools of 52% of the cases as contrasted to their absence in the stools of 116 control children. Subsequent studies have amply confirmed these observations. For example, of 1537 children admitted with diarrhea to the Childrens Hospital National Medical Center in Washington, DC from 1974 to 1982, rotaviruses were detected in the stools of 34.5%. The contribution of this virus to acute gastroenteritis in infants and children has varied some in different countries and different studies: in Canada 11.0%,[99] Japan 89%,[129] Venezuela 41.3%,[200] and the U.S. 89%.[117] The infection is also common in developing countries, and the mortality is higher because of the lack of treatment centers where fluid

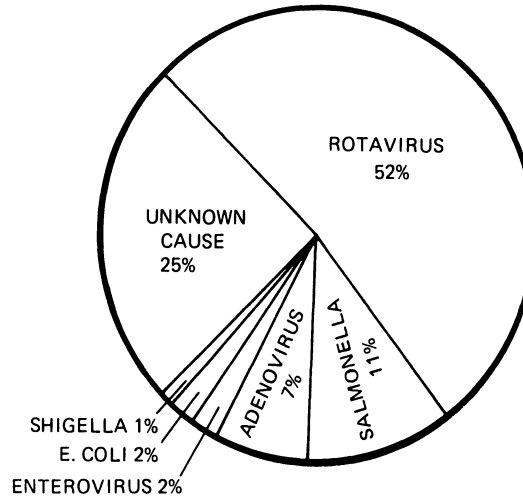

Figure 5. Causes of acute gastroenteritis in 378 children with enteric pathogens in feces. Adapted from Davidson *et al.* [46]

replacement or oral salts are available. In Bangladesh, rotaviruses were implicated in 46% of 6352 patients seeking treatment at the Matlab Treatment Center[22]; *E. coli* accounted for 28%, and bacterial pathogens predominated over 2 years of age.

12.2.6. Perinatal Infections. The acronym TORCH (toxoplasma, other viruses, rubella, cytomegalovirus, and herpes virus) was earlier used to designate the main causes,[60,180] but the recognition of HBV, HIV, and other viral causes has made the designation less useful. The estimated infection rate in pregnant women and their offspring, as derived from recent sources,[2,7] is presented in Table 6. Infections may be acquired *in utero* via the placenta, leading to congenital abnormalities, during passage through the birth canal, or shortly after birth in the neonatal period, often leading to long-term persistent infections.

Cytomegalovirus is the most common and important cause of congenital infections and abnormalities now that rubella vaccination has diminished this infection in pregnancy and in the contacts of pregnant women. Cytomegalovirus produces congenital anomolies such as microencephalopathy, chorioretinitis, deafness, and mental retardation, although it should be recognized that CMV infection of the mother is usually asymptomatic and benign; it is the large number of infections (about 2%/year in antibody-negative pregnant women) that raises CMV congenital abonormalities to importance. These are often the consequence of primary CMV infections but may occasionally result from reactivated infections. The prevalence of CMV in women of child-bearing age varies in different socioeconomic and geographic settings: in the United States 40–70% have antibody, leaving

Table 6. Estimated Risk of Infections in Pregnant Women and Their Offspring in the United States[a]

Cause	Mother (per 1000 pregnancies)	Fetus[b] (per 1000 live births)	Neonate[b] (per 1000 livebirths)
Viruses			
CMV			
During pregnancy	10–130	6–34	—
At delivery	30–280	—	20–100
HBV[c]			
Acute infection	Same as gen. population	—	50–100%
Antigen carriers	1–3	—	5–8%
HSV	1–10	Rare	<0.1–0.5
Rubella			
Epidemic	20–40	4–30	None
Interepidemic	0.1–1.0	0.2–0.5	None
VZV	0.5–0.7	Very rare	Very rare
Other causes			
T. gondii	1.5–6.4	0.75–1.3	—
T. pallidum	0.2	0.1	—

[a]Derived from Alford *et al.*[2] and Andiman and Horstmann.[7]
[b]Number of infections acquired by fetus *in utero* or by the neonate at birth or in the early postpartum period.
[c]Acute hepatitis: acquired during third trimester or within 2 months postpartum. The antigen carrier state refers to the percentage of exposed infants who become HBsAg carriers during first 6 months of life.

the remainder susceptible to primary infections, and 3–12% are excreting virus in the urine, as do about 1% of newborns (see Chapter 8, Section 5.1). In one prospective study of antibody-negative women, 1.4% had a primary CMV infection during pregnancy, leading to a 77% infection rate and 11% stigma rate in their offspring. In the United States and England, about 1 in 100 live births will be CMV infected, and about 1 in 1000 will have a congenital defect.

Rubella is the next most common congenital infection, resulting in abortion and stillbirth and in congenital abnormalities such as cataracts, deafness, heart lesions, and psychomotor retardation (see Chapter 23, Section 8.1). Congenital abnormalities occur in 15–20% of infants born of mothers infected during the first trimester; late manifestations may increase this rate to 30–45%.

Herpes simplex virus (HSV) infections of the pregnant mother with either type 1 or type 2 can lead to disseminated disease of the newborn, as can herpes zoster (VZ), with involvement of the liver and other visceral organs as well as localized lesions. The risk per 10,000 mothers is estimated as 10 for mumps, five for varicella zoster, and 0.5 for measles (rapidly lowering as measles vaccination extends to this young adult age group). Mumps and measles infections of the pregnant mother may result in increased fetal death, and VZ and vaccinia may result in widespread infections of the infant. Maternal Coxsackie B infections may lead to neonatal encephalomyocarditis. Hepatitis B (HBV) can infect *in utero* or at the time of birth, the latter being a very common method of transmission in developing countries, where it leads to persistent antigemia, cirrhosis, and an over 200-fold risk of hepatocellular cancer in young adults over those not infected; this tumor is more common in males. The data in Table 6 are for the United States; in the Far East rates of 50–200/10,000 pregnancies occur, and 40–73% of exposed infants become carriers of the virus in the first 6 months of life. Efforts are being carried out in Taiwan to prevent this perinatal infection with HBV vaccine and immune globulin in the hopes of preventing the later development of hepatic cancer. The human immunodeficiency virus, HIV, can also infect perinatally and *in utero* and results in AIDS and with EBV lymphocytic interstitial pneumonitis (LIP).

12.2.7. Immunosuppressed and Surgical Patients. Reactivation of viral infections, especially herpes, is common in immunosuppressed, transplanted, transfused, or HIV-infected patients. Most of these are inapparent infections, but diseases occasionally result, especially with CMV and with EBV infected. Armstrong *et al.*[9] found the infection rates in 26 prospectively followed renal-transplant recipients to be: CMV, 43%; HSV, 28%; EBV, 32%. With the exception of three primary CMV infections, all others represented reactivation. No unusual incidence of primary or secondary infections to nonherpes viruses (parainfluenza 1, 2, 3, HBV, measles, rubella) occurred. Clinically, five patients developed herpetic-type sores, three of whom showed HSV antibody rises; five had fever of unknown origin with rises in CMV antibody titer. Hematologically, seven patients showed atypical lymphocytosis associated with serological evidence of CMV in six. Of 13 episodes of rejection, five occurred in patients with CMV antibody rises. Fever and lymphocytosis caused by CMV also occur after cardiac surgery, and the mononucleosis syndrome occurs in about one-third of patients after heart surgery with an extracorporeal pump (see Chapter 8, Section 8.2.3). The source of CMV in transplant and surgery patients is unclear, and it might be exogenous in origin, be introduced with blood, result from reactivation in the blood of the recipient, or be present in the transplanted organ. Immunodeficiency also enhances the severity of primary viral infections, especially herpes infections, from natural exogenous sources as well as induced infections in persons receiving live polio, measles, rubella, smallpox, or yellow fever vaccines.

12.2.8. Sexually Transmitted Infections. These now include a wide variety of agents and both heterosexual and homosexual transmission. The viruses include CMV[207], genital herpes (including both HSV-1 and HSV-2), HBV, papilloma virus, HIV, and probably HILV-I. Only diseases resulting from HBV and HIV infections (AIDS) are currently reportable in the United States. It has been estimated that one case of genital herpes occurs for every five to ten of gonor-

rhea[156]; in a group of Boston clinics, genital herpes was seen seven times more frequently than primary syphilis.[31] The distribution of the reasons persons visited clinics for sexually transmissible diseases (STDs) in 12 cities in the United States from 1977 to 1985 is summarized in Table 7 (W. Jenkins, CDC, personal communication, 1986). Gonorrhea was the most common identified cause, accounting for 22.3% of visits in males and 20.6% in females. Nonspecific urethritis or vaginitis was diagnosed in 22.2% of males and 8.0% of females. Note that the extent to which *Chlamydia trachomatis* infections contributed to this syndrome was not reported. In males, genital herpes was diagnosed in 2.3% and in females in 1.6% of the visits. Trichomonal vaginitis was seen in 10.9% of female visits. In more intensive etiologic studies carried out by the STD clinic in Seattle (H. Handsfeld and K. K. Holmes, personal communication, 1986), a causal agent was identified in 5738 persons among some 25,000 attending the clinic for the first time. The distribution of known infections was gonorrhea in 28%, *C. trachomatis* infections in 28%, herpes in 20%, and human papilloma virus in 23%. Some 25% of the gonorrhea infections were followed by *C. trachomatis* infections. Among active male homosexuals in large urban centers, antibody prevalence rates to CMV, EBV, HBV, and HSV are extremely high, and HIV antibody prevalence rates are up to 60–70% in certain areas. The spread of HIV infections to wives, prostitutes, and other heterosexual partners (and often to the children of such partners), as well as to others via shared needles, transfusions, or other blood products, is a matter of great public-health concern.

12.2.9. Nosocomial Infections. Viruses are estimated to cause at least 5% of nosocomial infections,[134,205] but few systematic studies have been done because of the cost and time involved. Some 28 viruses have been documented as being nosocomially transmitted,[53] but the relative importance of few of these has been evaluated in the total nosocomial picture or even in special populations. Several groups of viruses have been implicated. These include respiratory viruses, especially influenza, parainfluenza, and RSV, the herpes group, especially CMV, HSV, and VZ, the hepatitis viruses including HAV, HBV, and non-A, non-B, gastroenteric viruses, mainly rotaviruses, the viruses of several exanthems such as rubella and measles, and occasionally the picornaviruses. Recent concern has centered on the nosocomial transmission of the AIDS virus to staff, but present clinical and serological evidence indicates a very low level of risk with the exception of a couple of infections resulting from needle sticks. A few instances of transmission of a slow virus (Creutzfeldt–Jakob) and of rabies virus have been documented under special circumstances such as corneal transplants. In the appropriate tropical setting, Lassa fever, Ebola virus, and Marburg virus have been transmitted as nosocomial infections. Lassa fever, in particular, has infected patients and staff, especially in the obstetrical wards via infected placentas.

Table 7. Percentage Distribution of Causes for Visits to Sexually Transmissible Disease (STD) Clinics in 12 Cities in the United States from 1977 to 1985[a]

Cause for visit	Males (322,422 visits)	Females (130,320 visits)
Gonorrhea	22.3	20.6
NSU or NSV	22.2	8.0
Venereal warts	3.6	3.3
Pediculosis pubis	3.4	2.5
Genital herpes	2.3	1.6
Syphilis	1.7	1.0
Scabies	1.0	0.4
Trichomonal vaginitis		10.9
Candida vaginitis		7.2
Visits for other purposes	41.6	41.9

[a]Derived from data kindly provided by Dr. William Jenkins, Statistics Branch, Centers for Disease Control, Atlanta, GA (personal communication).

One of the most carefully studied viruses is RSV, which appears to be the major nosocomial agent on some pediatrics wards, where it can infect both patients and staff; an outbreak among elderly patients in a health-care facility has also been reported.[101,102] Most studies of nosocomial infections have centered on certain wards or population groups. One of these is the pediatric population in the neonatal unit, the nursery, and the intensive care wards, where susceptibility, crowding, and often an immature immune system make these high-risk groups. In these settings, infections, diseases, and outbreaks of CMV, HSV, VZ, enteroviruses, myxoviruses, parainfluenza viruses, and especially RSV have occurred. Among neonates, RSV infections can be severe and atypical, with a high mortality. In one prospective study, 45% of exposed infants hospitalized for a week or longer acquired RSV infection, and it involved 42% of ward personnel.[101,102] Parainfluenza and rhinovirus infections may also be widely spread in these settings and may involve transmission by personal contact as well as by fomites, thus emphasizing the need for good handwashing techniques. Rubella has spread to other infants and to susceptible nursery staff; thus, vaccine protection of female staff of child-bearing age is of importance.

A second setting is the hemodialysis unit and laboratory, where staff are exposed by aerosol or blood to HBV, non-A, non-B hepatitis, and probably to HIV. A third setting is on any crowded ward or intensive care unit where both susceptible staff and patients are at risk for influenza and other respiratory agents. For this reason, routine influenza immunization should be carried out yearly for all hospital

yearly for all hospital personnel, not only for their own protection but also to prevent spread to patients. Fourth, patients receiving multiple transfusions or transplanted organs may be infected with the hepatitis viruses, CMV, HIV, and sometimes with EBV. Immunosuppressed patients are especially subject to both primary and reactivated infections, among which the herpes viruses are the most common.

12.2.10. Renal Syndromes. In renal diseases, evidence of viral causation has not been firmly established in man except for hemorrhagic cystitis, which is caused by adeno 11. This is despite the occasional presence of viruses in the urine and the ability of many viruses to multiply in *in vitro* tissue cultures prepared from human kidneys. The role of immune complex formation of viruses and antibody in the causation of human glomerulonephritis is unknown, although there is ample precedent in animal models[168]; except for elevated antibody titers to rubella virus in the nephritis of systemic lupus erythematosus, no other leads were found in a serological study of 106 cases of immune complex glomerulonephritis of unknown cause employing 13 different viral antigens.[210] It is likely that improved techniques of identifying viruses and immune complexes will lead to the discovery of a role for viruses in both acute and chronic nephritis.

13. Diagnosis of Viral Diseases

The etiologic diagnosis of a viral disease usually requires laboratory tests. There are four circumstances in which a probable diagnosis of the causative agent is suggested on clinical or epidemiologic grounds or both. First, some viral infections have distinctive enough clinical features that typical cases can be recognized if they occur in the right geographic area, season, and/or age group. This includes chickenpox and herpes zoster, herpes simplex infection of lips or genitalia, infectious mononucleosis in a young adult, measles, mumps parotitis, paralytic poliomyelitis, rabies, rubella, smallpox, and viral hepatitis. Second, if there is an epidemic in which an etiologic agent has been isolated, then most clinical syndromes of the same type are probably caused by the same virus. Examples of this are outbreaks of influenza, arbovirus infections, enteroviral exanthems, epidemic pleurodynia, and pharyngeal–conjunctival fever. Third, special or unique epidemiologic circumstances may indicate the probable diagnosis; croup or bronchiolitis in an infant is most likely caused by RSV, jaundice in drug users or homosexuals is usually hepatitis B or non-A, non-B hepatitis or following a blood transfusion is often non-A, non-B hepatitis, and mononucleosis following blood transfusion and/or immunosuppression is probably caused by CMV. Fourth, the type of organ involvement may be a lead—i.e., 80–90% of common respiratory infections are viral in origin, and nonpurulent infections of the CNS are likely to be viral, with the most likely candidates being HSV, mumps virus, enteroviruses, and arboviruses, in that order.

There are some common but not pathognomonic features of viral diseases: they are usually nonpurulent and associated with mononuclear rather than polymorphonuclear infiltrates; the onset is more likely to be insidious than with a bacterial infection; often there are prodromal symptoms; and retrobulbar headache is common. In the clinical laboratory, the presence of a normal or low white count suggests a viral infection, but typhoid, tuberculosis, brucellosis, malaria, histoplasmosis, and overwhelming bacterial infections can also produce leukopenia. The presence of lymphocytosis and of atypical lymphocytes also suggests a viral infection. Lymphocytosis of 50% or more and atypical lymphocytosis of 20% or more occur in infectious (EBV) mononucleosis, CMV and HIV mononucleosis, and rarely in *T. gondii* infections, but drugs such as *p*-aminosalicylate (PAS), phenytoin, and tetrachlorethylene may also evoke lymphocytosis. Less intense lymphocyte responses are seen in a variety of viral infections such as rubella, hepatitis A, adenovirus, mumps, herpes, and varicella infections. They may occasionally occur in tuberculosis, histoplasmosis, and other nonviral infections. Evidence of a reversal of the $T_4 : T_8$ lymphocyte ratio or low absolute T_4 counts points to HIV infection; the degree of change reflects the risk of clinical disease: A decreased T_4 suggests HTLV-I.

The diagnostic procedures used for viral infections are presented in individual chapters of this book. However, there are certain common aspects of collection, requests for testing, and interpretation that merit comment here. A detailed description can be found in the APHA diagnostic handbook[135] and in a fine book by Hsiung.[110]

13.1. Collection

Materials for viral isolation should be obtained from the site of the lesion, if feasible. Usually, a swab or gargles from the throat and a rectal swab or a stool sample are useful for all suspected respiratory and CNS infections and for the viral exanthems. Nasopharyngeal washings using an infant-sized catheter and suction apparatus should be taken for direct fluorescent-antibody or ELISA test for identification of respiratory viruses.[92,180] Amplification of the amount of virus prior to these tests by growth in suitable tissue cultures for 24–48 hr increases the sensitivity.[80] In suspected arbovirus infections, a sample of whole blood should be collected, and in a vesicular exanthem, an aspiration or scraping of the lesion. Direct viral fluorescent-antibody identification of rabies and certain other infections may be possible with small skin biopsies; brain biopsies are needed to identify herpes

simplex virus meningoencephalitis. All such materials should be frozen immediately at −70°C and shipped in dry ice or liquid nitrogen to the nearest viral diagnostic laboratory—usually a governmental (state, Federal) or university laboratory. Collection and shipping kits are often available. For some more stable viruses, freezing may not be necessary if transportation time is short. Certain laboratories now provide tubes with a transport medium. These include tissue-culture tubes with one or even two tissue-culture cell types already grown and ready for bedside inoculation of the specimen and shipment to the laboratory for further study.[149]

Serological tests are carried out on serum from a sample of blood, usually 10 ml collected in the acute illness, and on a convalescent sample obtained 2–3 weeks later; a third sample drawn about a month after the second may be useful in some infections. The sera should be sterilely separated immediately after clotting and either frozen (−20° or −70°C) or kept at 4°C. Serum may be stored in a freezer in the hospital or clinic where it has been collected or in the laboratory where the test is performed. In infectious-disease hospitals or units, routine collection and storage of acute and convalescent sera from all febrile patients should be carried out to permit retrospective testing.

13.2. Requests for Testing

Most common clinical syndromes have more than one cause, so a request for a battery of serological tests should be made for most individual cases. The laboratory needs clinical and epidemiologic information as a guide for these determinations. At a minimum, the age of the patient, date of onset, and the major organ system involved should be indicated on the request slip. The term "viral disease" or "FUO" leaves the laboratory at a loss as to the best way to proceed.

13.3. Tests Employed

Isolation or identification of the virus, preferably from the lesion itself, and demonstration of a serological response to it are the common criteria for viral diagnosis. Unfortunately, the isolation of a virus in tissue culture, embryonated eggs, or suckling mice may require at least a week and often longer if identification of the virus is involved, and serological diagnosis is usually dependent on tests of a convalescent serum sample. Such viral diagnosis is therefore of little help to the clinician in recognition and management of the acute viral disease. Progress is being made in rapid diagnostic techniques.[75,197,213] These include the identification of respiratory syncytial, influenza, parainfluenza, corona, and adenovirus antigens by fluorescent microscopy or ELISA tests[83] in nasopharyngeal cells[92]; the demonstration of rotaviruses, parvoviruses, and hepatitis A virus particles in stool samples by immune electron microscopy or by ELISA[146,197,213]; and the recognition of several viruses in clinical material by radioimmunoassay. The electron microscope and immunofluorescent (IF) techniques have also been used to identify herpes and pox viruses in vesicle fluid and CMV in urine. The demonstration of virus-specific IgM in the acute-phase serum with the ELISA or IF methods also permits early diagnosis. These rapid diagnostic techniques have been reviewed by WHO.[213,215]

13.4. Interpretation of Tests

Rapid identification of the virus in infected secretions or tissues or isolation of the virus and a fourfold or greater rise in antibody titer between the acute and convalescent sera are classic criteria for viral diagnosis. For some viral infections, isolation of the virus first and then tests for a serological rise against that isolate are required. This is true of virus groups in which there are too many antigenically distinct strains to carry out a battery of serological tests such as the echovirus, coxsackievirus, and rhinovirus groups. The adenoviruses, group B arboviruses, and influenza A and B groups have common intragroup antigens, particularly in the complement-fixation test. These permit one test to be used to indicate infection for all members of that viral group.

If it has been possible to obtain only a single convalescent serum sample and a high antibody titer is found, or if high titers are present in both acute and convalescent sera without a fourfold difference, then the question is whether these results reflect current infection or persistently high titers from a previous infection. Significance may be attached to these findings if the disease is a rare one in which the presence of this antibody is unique, if the test reflects a short-lasting antibody, or if IgM-type antibody can be demonstrated. A rapid drop in antibody titer in a subsequent specimen is also suggestive of a recent infection. Sequential testing of other family members may also be useful, since they may be in different stages of apparent or inapparent infection with the same virus. In an epidemic setting, comparison of the geometric mean antibody titer of sera collected early in illness from one group of patients with the titer in sera from another group of patients convalescing from the same illness may permit rapid identification of the outbreak.

Sometimes a virus may be isolated or an antibody rise may be demonstrated that is not, in fact, causally related to the illness. Sometimes two viruses, or a virus and a bacteria, are implicated in the infection, and the interpretation of their causal role may be very difficult. On other occasions, no virus can be isolated, or a serological rise is not demonstrable when a specific virus is the real cause of the illness. A list of some common causes for these false-positive and false-negative results is given in Table 8.

Table 8. Viral Diagnosis: Some Causes of False-Positive and False-Negative Tests

False positive

Viral isolation

1. Persistent or reactivated virus from prior and unrelated infection has been isolated.
2. A viral contaminant is present in the tissue culture or other isolation system.
3. Nonspecific cytopathic effects occur because of toxicity of specimen or presence of bacteria or other causes and are mistaken for a virus.
4. Two microbial agents are present, and the one isolated is not the cause of the disease.

Serological rise

1. Cross-reacting antigens.
2. Nonspecific inhibitors.
3. Double infection with only one agent producing the illness.
4. Rise to vaccination rather than natural infection.

False negative

Viral isolation

1. Viral specimen taken too late or too early in illness.
2. Wrong site of multiplication sampled (e.g., throat rather than rectal swab).
3. Improper transport or storage of specimen—not kept frozen.
4. Wrong laboratory animal or tissue-culture system selected for isolation.
5. Toxicity of specimen kills the tissue culture, obscuring the presence of virus.

Serological rise

1. Specimens not taken at proper time—i.e., too late in illness or too close together to show antibody rise.
2. Poor antibody response—low antigenicity of the virus or removal of antibody by immune-complex formation.
3. Wrong virus or wrong virus strain used in the test.
4. Nonspecific inhibitor obscures true antibody rise.
5. Wrong test used for the timing of the serum specimens.

14. Proof of Causation

The classic concepts of causation in infectious diseases are those elaborated by Jakob Henle (1809–1885) in 1840 and by his student Robert Koch (1843–1910) in 1884 and 1890. These are termed the Henle–Koch postulates. The basic criteria (Table 9, column 1) included the consistent presence of the parasite in the disease in question under circumstances that can account for the pathological changes and clinical course, the absence of the parasite in other diseases as a fortuitous or nonpathogenic parasite, and the experimental reproduction of the disease by the organism after having been grown repeatedly in pure culture. The inability of many clear-cut causes of certain diseases to fulfill these criteria was recognized by Koch himself and other limitations were later recognized.[65] He recognized that whereas the bacteria of anthrax, tuberculosis, tetanus, and many animal diseases fulfilled the proof, those of many other diseases did not. These latter included typhoid fever, diphtheria, leprosy, relapsing fever, and Asiatic cholera. He felt particularly strongly about cholera because he himself had discovered the causative organism. For these diseases, he felt that fulfillment of only the first two criteria was needed and that experimental reproduction of the disease was not essential to proof of causation. Rivers[181] reviewed the Koch postulates in terms of viral infections in his presidential address to the American Immunological Society in 1937 and found them lacking. Included in his objections were (1) the idea that a disease is necessarily caused by only one agent, citing the work of Shope[191] with swine influenza, in which both a virus and a bacteria are required; (2) the necessity of demonstrating the presence of viruses in *every* case of the disease produced by it; and (3) the fact that the existence of virus carriers must be recognized. He set forth two conditions for establishing the specific relationship of a virus to a disease (Table 9, column 2): (1) a specific virus must be present with a degree of regularity in association with the disease, and (2) the virus must occur in the sick individual not as an incidental or accidental finding but as a cause of the disease. In support of the latter, he stressed the importance of the experimental reproduction of the disease in susceptible experimental hosts with the inclusion of suitable controls to eliminate the fortuitous presence of other viral agents either in the patient or in the experimental host. The absence of antibody to a virus in the patient's sera at the onset of illness and its appearance during recovery were recognized as an important but not absolute link in causation; Rivers was cautious in this statement because of the possible presence of passenger viruses to which antibody appeared but that were not of etiologic significance. He also noted that recovery from viral infection sometimes takes place without the development of antibodies and that occasionally an individual already possessing antibodies against a virus succumbs to a disease caused by it (i.e., reinfection or reactivation).

The "virologists' dilemma" was further discussed in 1957 by Huebner,[111] who revised the Koch and Rivers postulates into the following criteria: (1) the virus must be a "real entity," i.e., well established on animal or tissue culture passage in the laboratory; (2) the virus must originate in human tissues and be repeatedly present therein and not in the experimental animals, cells, or the media used to grow it; (3) the agent should be characterized early to permit differentiation from other agents, including immunologic comparisons; (4) the virus should have a constant association with the clinical entity in question; (5) the clinical syndrome should be experimentally reproducible in volunteers inoculated with the agent in a "double-blind" study; (6) carefully conceived epidemiologic cross-sectional and longitudinal

Table 9. Postulates of Causation

Bacteria[a] Henle (1840); Koch (1890)	Viruses[b] Rivers (1937)	Viruses[c] Immunologic proof (1973)
1. Parasite occurs in every case of the disease in question and under circumstances that can account for the pathological changes and clinical course of the disease.	1. A specific virus must be found associated with a disease with a degree of regularity.	1. Virus-specific antibody is regularly absent prior to illness.
2. Occurs in no other disease as fortuitous and nonpathogenic parasite.	2. Virus occurs in the sick individual not as incidental or accidental finding but as cause of the disease.	2. Antibody regularly appears during illness, including: a. Transient viral-specific IgM antibody b. Persistent IgG antibody c. Local antibody (IgA) at site of primary multiplication.
3. After being fully isolated from the body and repeatedly grown in pure culture, can induce the disease anew.	3. Transmissible infection is produced with a degree of regularity in susceptible experimental hosts by means of inoculation of material, free from ordinary microbes or rickettsiae, obtained from patients with the disease, and proper control and immunological studies demonstrate that the virus was neither fortuitously present in the patient nor accidentally picked up in the experimental animals.	3. Antibody production is accompanied by presence of viruses in appropriate tissues.
Only 1 and 2 were regarded as essential by Koch.		4. Absence of IgG antibody indicates susceptibility to the disease. 5. Presence of IgG antibody indicates immunity to the disease. 6. No other virus or antibody is similarly associated. 7. Production of the antibody (immunization) prevents the disease.

[a]Koch[127] (see Rivers[181]).
[b]Rivers.[181]
[c]Derived from Rivers[181] and Evans.[63,64]

studies are indispensable in establishing the role of highly prevalent viruses in human diseases; (7) the disease should be prevented by a specific vaccine. He also added an eighth consideration—financial support—which is so needed to carry out the virological and epidemiologic analyses required in establishing proof of causation.

The problem of establishing causality for viral infections has been exemplified by the relationship of EBV to infectious mononucleosis. In the beginning, no method of virus isolation existed, no susceptible laboratory animal was known, and EBV antibody was already present at the time the patient with infectious mononucleosis was first seen by the physician. The proof of causation had to rest on prospective serological investigations that fulfilled certain immunologic criteria.[61,106,160,186,202] The most important of these were the regular absence of antibody prior to disease, its regular appearance during illness, and the relationship of antibody to susceptibility and immunity[78,186] (see Table 9, column 3). To date, a vaccine has not been developed to prevent the disease. Advances in viral technology later permitted the identification of the presence and persistence of EBV in the pharynx of patients having acute infectious mononucleosis. Human and monkey transmission experiments with EBV have resulted in the reproduction of some but not all of the features of the disease (see Chapter 10). The web of causation is now firm that EBV causes all heterophil-antibody-positive infectious mononucleosis and most heterophil-negative cases.[62]

Similar seroepidemiologic techniques have been needed in studying the spectrum of infections produced by

hepatitis B antigen (HBAg) because of the difficulty of isolating the virus in the laboratory and the lack of a good experimental animal[24] (see Chapter 13).

The most difficult and challenging problems of causation are arising in establishing the possible relationship between certain viruses and various malignant and chronic diseases. In the former category is the relationship between EBV and Burkitt lymphoma, nasopharyngeal carcinoma,[59,62] and to lesser extent Hodgkin disease[73,76,77]; of HBV to hepatocellular cancer[12]; of genital herpes and papilloma virus to cervical cancer; and of human T-cell leukemia/lymphoma virus (HTLV-1) to adult T-cell leukemia. In the field of chronic diseases, the importance of slow or unconventional viruses in causing kuru and Creutzfeldt–Jakob disease, fatal infections of the central nervous system, has been well established, as have the causal relationship of measles virus to SSPE[44] and of papova virus to progressive multifocal leukoencephalopathy (see Chapter 30). Multiple sclerosis remains a mystery.[43,113,188]

Much more tenuous is the possible viral causation of chronic diseases such as insulin-dependent diabetes mellitus, rheumatoid arthritis, systemic lupus erythematosus, and sarcoidosis. High antibody titers to EBV and certain other viruses have been found in about 40% of sera from cases of sarcoidosis[28] and systemic lupus erythematosus,[177,183] but these are probably secondary to polyclonal B-cell proliferation rather than representing a causal relationship. In contrast, in Burkitt lymphoma it has been clearly shown that high EBV-VCA IgG antibody elevations precede the development of the tumor such that a twofold titer elevation above normal constitutes a 30-fold risk for the tumor compared with children with normal levels.[48] The virus has also been consistently demonstrated in tumor tissue, and a malignant tumor has been reproduced in nonhuman primates, as discussed in Chapter 26.

High EBV antibody levels have also been shown to precede the diagnosis of Hodgkin disease in a pilot study of two cases,[73] and a large prospective study of 44 cases and matched controls has confirmed the increased risk of Hodgkin disease in the presence of elevated EBV antibody titers.[154] However, the role of the virus in tumor causation, if any, is probably an indirect one, since the virus or its genomes have rarely been found in tumor tissue.[173] Prospective studies of the relationship of HBV to hepatocellular cancer in Taiwan have clearly established the presence of HBVsAg many years prior to the tumor, with a 223-fold increased risk of the cancer in those with antigenemia over those without.[12] This virus–tumor causal association is discussed in Chapter 29 and represents the strongest current proof that a virus can cause human cancer. This will be firmly established if the ongoing trials of HBV vaccine in infants can prevent the development of the tumor in young adult life; this will take many years to determine, since the infection occurs in infancy and the tumor in young adult life.

The persistence and/or reactivation of these viruses under circumstances of impaired cell-mediated immunity (CMI) have been postulated as a possible common mechanism.[58,203] Such an impairment in CMI could arise when the viral infection occurs very early in infancy or during pregnancy; it might also result from the presence of a concomitant infection (malaria) that depresses the immune response, from the use of immunosuppressive drugs, from genetic defects in the ability of T-type lymphocytes to recognize or respond to certain viruses, from serum inhibitors of cellular immunity, or from disease-induced immunosuppression (Hodgkin disease, HIV infections).

Current evidence thus suggests that certain cancers and certain chronic diseases of man are caused by the persistence and/or reactivation of common, ubiquitous viruses in an immunologically compromised host. Those viruses with a capacity for latency such as the herpes, papova, measles, rubella, and adenoviruses appear to be the most likely candidates for the causation of these conditions. Present and future work to determine the elements of causation include (1) large-scale multipurpose prospective studies of populations, seeking evidence of viral persistence, high viral antibody levels, and/or impaired lymphocyte response to viral agents as a possible prelude to malignancy and chronic disease, and then the appearance of the disease itself as more definitive proof of causation; (2) the demonstration of the virus or viral genome in afflicted tissues but not in normal tissues; (3) the occurrence or reproduction of the condition in man and/or experimental hosts, or both, under natural or induced viral infection. It must be stressed that cancer or a chronic disease will not always result even under propitious circumstances. The host response will probably fall along a biological gradient from very mild to severe.

It also seems likely that any given malignant or chronic condition may be produced by more than one cause or group of causes. The current evidence on viruses, cancer, and their relationship to chronic neurological diseases is discussed in later chapters of this book. The developments in our concepts of causation and the limitations of the Henle–Koch postulates have recently been reviewed.[63–65] A unified set of guidelines has been proposed for both infectious and noninfectious diseases.[63] However, existing postulates concentrate on the relationship between a suspected cause and the resulting clinical illness. Yet most viral infections result in many subclinical or inapparent infections for every one that is clinically manifest. Subclinical illness is also common in bacterial infections as well as in many chronic diseases such as coronary heart disease, diabetes, and some malignancies.

Once the pathogenic process has been initiated, some additional factor or factors may be needed to result in clinical

illness. These have been termed "clinical illness promotion factors."[70] In infectious diseases, these factors are incompletely understood and vary from one disease to another. For some, the age at the time of infection is an important determinant (poliomyelitis, viral hepatitis, infectious mononucleosis); for others, genetic susceptibility to the infection and/or the disease among those infected probably plays an important role, perhaps operating through the immune system, as in the X-linked lymphoproliferative syndrome associated with EBV.[179] Psychosocial factors have also been shown to be important in the development of infectious mononucleosis among those infected with EBV.[119] Focus on the means of preventing the emergence of clinical illness among those infected is of special relevance to a virus like HIV because over a million and a half persons are currently infected in the United States, of whom some 50% or more will develop AIDS or a related illness with a case mortality of over 70%.

15. Control and Prevention

The basic concept in controlling a viral disease is to break a link in the chain of causation. Interruption of a single known essential link may effectively control a disease even if knowledge of other links, or of the etiology itself, is incomplete. Despite this, very little has been accomplished in most viral diseases by environmental changes except for the arboviruses, in which the appropriate insect vector can be controlled. Improved water supplies, proper sewage disposal, and improved personal hygiene could potentially decrease the incidence of poliomyelitis and other enterovirus and hepatitis A infections, but in general the results have been disappointing because so many pathways of infection exist. Furthermore, improved sanitation may delay the age of exposure to later childhood and young adult life, when infections are more often clinically apparent and more severe.

15.1. Immunization

The difficulty in the environmental control of viral infections spread by close personal contact, by the respiratory route, or even by oral–intestinal spread has directed the main thrust of prevention to immunization of the host. The requirements of a good vaccine are listed in Table 10. The overall objective is to create the same degree and duration of protection as with natural infection but without the accompanying clinical illness. Both live and killed vaccines have been used. A comparison of live and killed vaccines is given in Table 11. In general, live viral vaccines are more desirable and induce a longer and broader immune response, especially if given by a natural route. Some of the problems include successful attenuation without reversion to virulence, avoidance of viral persistence and the risk of reactivation, and the elimination of possible oncogenicity. These are major hurdles for vaccines against herpes viruses, and it is difficult to measure some of these attributes in the labora-

Table 10. Objectives of Immunization

1. Produce a good humoral, cellular, and local immune response similar to natural infection.
2. Produce protection against clinical disease and reinfection.
3. Give protection over several years, preferably a lifetime.
4. Result in minimal immediate side reactions or mild disease and with no delayed effects such as late reactivation, CNS involvement, or cancer.
5. Can be administered simply in a form acceptable to the public.
6. Cost and benefits of administration should clearly outweigh the cost and risk of natural disease.

Table 11. Comparison of Live and Killed Vaccines[a]

	Live	Killed
Immune response		
Humoral antibody (IgG)	+++	+++
Local antibody (IgA)	+++	+
Cell-mediated immunity	+++	+
Duration of response	Long	Shorter
Epidemiologic response		
Prevents reinfection by natural route	+++	+
Stops spread of "wild" virus to others	+++	+
Some vaccine viruses (polio) spread to others	+++	0
Creates herd immunity if enough persons are vaccinated	+++	0
Characteristics of the vaccine		
Usually heat stable	0	++
Vaccine virus may mutate or increase in virulence	+	0
Antigenic loss possible during preparation (e.g., formalin treatment)	0	+
Contraindicated in immunosuppressed persons	+++	0
Side reactions: systemic (viremia)	+	0
local	0	++
Number of doses for successful take	1[b]	2–3

[a]The table is a simplification and may not apply to all vaccines. Some live vaccines (polio) are relatively heat stable. The induction of local immunity is often dependent on the antigenic dose of killed vaccine; some induce sufficient immunity to lower reinfection rates and decrease spread of wild virus. Our knowledge of the presence and degree of cell-mediated immunity is inadequate for many vaccines.

[b]Several doses of polio vaccine are given to insure a take against the three types on at least one of these.

tory. There are efforts to produce live vaccines with temperature-sensitive mutants for respiratory syncytial and influenza viruses that would multiply only in the colder temperature of the upper respiratory host but not in the lung where clinical disease might result. Table 12 summarizes current information on the use of viral vaccines, and Table 13 summarizes recent recommendations for normal children and infants. WHO lists requirements for international travel.[217]

The most successful of these efforts toward vaccine development have used an attenuated live virus as the antigen (adenovirus, measles, mumps, poliovirus, rubella, and smallpox). Administration by the natural portal of entry to produce local immunity has also been important (poliovirus, adenovirus). Inactivated viral vaccines such as influenza vaccine have met with limited success, although highly purified and concentrated preparations are giving more promising results.[82] Killed poliovaccine has been successfully employed as the sole method of vaccination in several countries such as Sweden, Finland, and The Netherlands in the past, but some problems arising for religious reasons led to an outbreak in unimmunized persons that spread to Canada and the United States.[33] In Finland, waning immunity was apparently the reason for an outbreak in which oral vaccine was added to the program. On the other hand, a new, potent killed vaccine has been shown to produce high seroconversion rates after one or two injections and has been field tested in Senegal in combination with DPT in two injections 6 months apart.[194] It is useful in areas where the response to oral polio vaccine has been poor, in highly endemic areas where mass oral programs are difficult, in immunocompromised persons, or in susceptibles exposed to OPV. Use of both may be useful in some areas.

Passive immunization is a short-term expedient useful only when the γ-globulin can be administered soon after exposure and when it contains a sufficiently high titer of specific antibody against the agent. In some instances, preparations are derived from persons known to be convalescent from the disease, from persons hyperimmunized against it, or by selecting only donors shown to have high antibody titers. Today, passive immunization is generally limited only to well-defined exposures to HAV, vaccinia virus (unlikely now that immunization has been discontinued throughout the world but which might again arise if vaccinia virus is used as the carrier for other antigens), and rabies virus under circumstances of high exposure or high susceptibility in the host (immunocompromised persons). In rabies this approach is important early in severe exposures, as it may limit local multiplication and subsequent spread of the virus to the CNS.

The rapidly expanding knowledge of molecular virology, of DNA technology, and of the function and cloning of various parts of the genome of viruses, of their insertion into carrier vehicles, and of the concept of preparing idiotypic vaccines directed against the receptor for the virus on the host cell have led to a plethora of new experimental vaccines that offer great hope for the future. To facilitate the further development of these methods, the National Institute of Allergy and Infectious Diseases (NIAID), National Institutes of Health, of the United States initiated a priority list in cooperation with the Institute of Medicine (IOM) in 1982.[158] Ten diseases or disease categories caused by 17 different organisms were targeted for new or improved vaccines within the next 10 years. In a domestic model, the following five vaccines were assigned the highest priority in the order listed: hepatitis B virus (HBV, recombinant DNA derived), respiratory syncytial virus (RSV, live attenuated), one bacterial vaccine *Hemophilus influenzae*, type b (hib), influenza (subunit), and varicella virus (immunocompromised children). In an international model, five vaccines were recommended for development, among which was one viral agent, rotavirus, assigned the second-highest priority.

As of mid-1988, encouraging progress has been made in the understanding of the biology of all these agents as well as in the vaccines themselves. A live attenuated varicella vaccine for children with acute lymphocytic leukemia is nearing licensure, based on the earlier work of the Japanese, and a license is approved for a recombinant-DNA-derived hepatitis vaccine with the antigens expressed in yeast or mammalian cells to replace the plasma-derived hepatitis surface antigen in inactivated HBV vaccines. Live attenuated influenza and rotavirus vaccines are undergoing evaluation in field trials, and a live attenuated hepatitis virus (HAV) is undergoing initial studies in humans. Progress on the development of RSV is being made now that the complete nucleotide sequences for the major glycoprotein (G) and fusion protein (F) have been determined and that two separate serotypes of the virus circulating both at the same and separate times have been recognized. A vaccinia virus recombinant containing the G gene is under test in rats. A candidate vaccine consisting of purified HN and F glycoproteins for parainfluenza type 3 is under evaluation in animals, and cold-adapted mutants are also under study. Clinical trials of a cold-adapted influenza A vaccine are in progress with special focus on whether cell-mediated immunity is induced, whether the vaccine can interrupt spread of the virus in a family settings, and whether appropriate antibody responses can be induced in infants. Improved vaccines for influenza B are progressing. Many approaches are being explored in the development of a rotavirus vaccine. One is the attenuation of the virus by serial passage in gnotobiotic piglets and tissue culture, a second the use of cross-reacting animal rotavirus strains, a third the use of an avirulent strain isolated from a rhesus monkey or one isolated from a calf, and a fourth the insertion of recently obtained cloned rotavirus genes, which are incorporated into a prokaryotic expression vector (*E. coli* K12) or into vaccinia virus to produce a vaccinia–rotavirus recombinant strain.

Indeed, the large vaccinia virus is being explored as a

Table 12. Viral Prophylaxis[a]

Vaccine	Type	Persons given to	Age	Product	Dosage	Route	No. of doses
Adeno.	L	Military recruits	Young adults	Types 4 and 7	0.5	Oral	1
HAV	IG	Household, sex contacts of cases	Any		0.02 ml/kg in first 2 weeks of exposure	I.M.	1
		Contacts in day care centers					
		Travelers to high-risk areas outside the tourist routes			0.02 ml/kg for 2–3 mo.	I.M.	1
					0.06 ml/kg for 5 mo.		
HBV	I	Persons at high risk: occupational, sexual, environ., travel, I.V. drugs, family, prisoners	Adults	HB	Two doses 4 wk. apart; third 5 mo. after second	I.M.	3
	IG	Exposure to HBV+ blood; sex or bite from case or carrier	Any	HBIG	0.06 ml/kg + one dose 1 mo. later except when HB is given	I.M.	2
Influenza A and B	I	High-risk conditions;	6–35 mo.	Split	0.25 ml	I.M.	2[b]
		nursing homes, certain chr. dis., health personnel, persons >65 years	3–12 yr.	Split	0.5 ml	I.M.	2[b]
			>12 yr.	Whole or split	0.5 ml	I.M.	1
A only	AV	Outbreaks in closed groups	Any	Amantadine	200 mg daily or 100 mg b.i.d. for > 65	Oral	Daily
		High risk: health personnel, immune-deficient persons					
Measles	L	Developed countries	15 mo.	MMR	One	S.C.	1
		Developing countries	9 mo.				
	IG	Exposed susceptibles	Any	IG	0.25 ml per kg (up to 15 ml) <6 days of exp.	I.M.	1
Mumps	L	All at risk	15 mo.	MMR	One	S.C.	1
Polio	L	All at risk	2, 4, 6, 18 mo., 4–6 yr.	OPV	One	Oral	5
	K	Immunocomp. persons, unimmunized travelers to endemic areas	Any	IPV	Three doses 4 wk. apart + fourth 6 mo. after third	S.C.	3
Rabies	K	Preexposure or high risk	Any	HDCV	1 ml at 0, 7, 28 days	I.M.	3
		Postexposure to wild animals, rabid or suspected rabid dog or cat, escaped animal	Any	HDCV	1 ml at 0, 3, 7, 14, and 25 days	I.M.	5
	IG		Any	HBIG (+ HDCV)	20 IU/kg	I.M.	1 + local
Rubella	L	All	15 mo.	MMR	One	S.C.	1
		All adults lacking documented vaccination or	Adults				

(*continued*)

Table 12. *(Continued)*

Vaccine	Type	Persons given to	Age	Product	Dosage	Route	No. of doses
		antibody neg., esp. women of childbearing age					
Smallpox		No indication for use in civilian populations					
		Lab. workers with virus					
V–Z	L	Leukemia	Children	Not yet available			
	IG	Immunocompromised and susceptible persons exposed to household or hospital cases	Any	VZIG	<50 kg, 125 U/10 kg >50 kg, 625 units	I.M.	
Yellow fever	L	Travelers or residents of endemic areas	>6 mo.	17-D	One dose >6 days before travel	S.C.	1

[a]Derived from references 32, 34–37. L, live vaccine; I, inactivated vaccine; IG, immune globulin; AV, antiviral drug; MMR, measles/mumps/rubella vaccine; HB, hepatitis B vaccine; HBIG, hepatitis immune γ-globulin; HDCV, human diploid cell vaccine; VZIG, varicella immune EGG-globulin.
[b]For influenza the two doses are given at least 4 weeks apart.

carrier of several other vaccines as well, including hepatitis, influenza, genital herpes, and a malarial antigen. A vaccine against HIV has been assigned a separate high priority but progress is very slow.

Although the major emphasis of these development programs is on live attenuated vaccine, several improved inactivated vaccines are also under study, such as influenza and hepatitis A viruses. An improved killed poliomyelitis vaccine has been developed in The Netherlands and has been successfully field tested in Africa both in separate vaccine trials and combined with DPT.[194] Despite these exciting new developments, it must be remembered that each new vaccine must be evaluated in carefully conducted field trials to prove that its efficacy, safety, cost, ease of administration, thermostability, and freedom from long- and short-term reactions or vaccine complications are better than those of existing vaccines. This will not be an easy task and in some instances may be an impossible one. Our greatest problem on a worldwide basis today is not the lack of efficacy of most available vaccines but in delivering them in a viable state and at an appropriate age to susceptible children before natural infection occurs.

15.1.1. Immunization in Developing Countries. The effective utilization of current vaccines, especially against childhood diseases, in tropical and developing countries presents many biological, economic, logistic, and political problems. Some of these are listed in Table 14. The need to initiate immunization in the very short period before natural infection occurs in these settings, the need for maintenance of the viability of live vaccines through an effective cold chain, the difficulty in transportation of vaccine to remote areas or during the rainy season or finding adequate health personnel to administer it on arrival, and the poor socioeconomic and educational levels in many settings are but a few of the difficulties. Despite these problems, the World Health Organization (WHO) has initiated an expanded program of immunization (EPI) directed at achieving immunization of all children in the developing world against six targeted diseases by 1990 as part of their development of primary care programs. The diseases are measles, poliomyelitis, diphtheria, pertussis, tetanus, and tuberculosis in children. It also includes immunization of mothers to protect against neonatal tetanus. Methods of surveillance of households to determine the need and use of the vaccines have been developed. This ambitious effort was amplified and expanded in 1984 through a cooperative effort sponsored by WHO, UNICEF, the World Bank, the United Nations Development Program (UNDP), and the Rockefeller Foundation to Create a Task Force for Child Survival.[216] The Task Force objective is to promote the reduction of childhood morbidity and mortality via the acceleration of key primary health care activities.

15.1.2. Eradication versus Control. The successful global eradication program against smallpox through the efforts of WHO has been a singular achievement in preventive medicine and has raised the hope that other diseases might be similarly controlled. The term has been characterized as follows: "Eradication of an infection implies that the infection has disappeared from all countries of the world because transmission of the causative organism has ceased in an irreversible manner."[196] It involves the control of the clinical disease with its attendant morbidity, disability, and

Table 13. New Recommended Schedule for Active Immunization of Normal Infants and Children

Recommended age[a]	Vaccine[b]	Comments
2 months	DTP-1,[c] OPV-1[d]	Can be given earlier in areas of high endemicity
4 months	DTP-2, OPV-2	6-week to 2-month interval desired between OPV doses to avoid interference
6 months	DTP-3	An additional dose of OPV at this time is optional for use in areas with a high risk of polio exposure
15 months[e]	MMR,[f] DTP-4, OPV-3	Completion of primary series of DTP and OPV
24 months	HbPV[g]	Can be given at 18–23 months for children in groups who are thought to be at increased risk of disease, e.g., day-care-center attendees
4–6 years[h]	DTP-5, OPV-4	Preferably at or before school entry
14–16 years	Td[i]	Repeat every 10 years throughout life

[a]These recommended ages should not be construed as absolute; i.e., 2 months can be 6–10 weeks, etc. From ref. 38.
[b]For all products used, consult manufacturer's package enclosure for instructions for storage, handling, and administration. Immunobiologics prepared by different manufacturers may vary, and those of the same manufacturer may change from time to time. The package insert should be followed for a specific product.
[c]DTP, diphtheria and tetanus toxoids and pertussis vaccine adsorbed.
[d]OPV, poliovirus vaccine live oral; contains poliovirus strains types 1, 2, and 3.
[e]Provided at least 6 months has elapsed since DTP-3 or, if fewer than three DTPs have been received, at least 6 weeks since last previous dose of DTP or OPV. MMR vaccine should not be delayed just to allow simultaneous administration with DTP and OPV. Administering MMR at 15 months and DTP-4 and OPV-3 at 18 months continues to be an acceptable alternative.
[f]MMR, measles, mumps, and rubella virus vaccine, live.
[g]*Hemophilus* b polysaccharide vaccine.
[h]Up to the seventh birthday.
[i]Td, tetanus and diphtheria toxoids adsorbed (for adult use); contains the same dose of tetanus toxoid as DTP or DT and a reduced dose of diphtheria toxoid.

Table 14. Immunization Problems in Developing Countries

1. Inadequate surveillance of infectious diseases.
2. Inadequate diagnostic facilities.
3. Inadequate and unreliable transportation, maintenance problems in the tropical environment, and the difficulties of movement in the rainy season.
4. Inadequate health personnel for surveillance, diagnosis, and the delivery of vaccines.
5. Inadequate funds for immunization programs.
6. Remote and dispersed populations in many areas.
7. Problems in record keeping because of illiteracy rate.
8. Problems in communication.
9. Problems in maintaining the cold chain for vaccines and lack of sufficiently heat-stable preparations.
10. Poor antibody response to some vaccines such as OPV because of poor nutrition, poor immune response, presence of inhibitors(?), interference by other agents, loss of antigenicity in tropical areas, inadequate dose because of faulty equipment and other unknown reasons.
11. Early age of infection, requiring immunization in the first year of life, perhaps earlier, even at the time of birth.
12. Difficulty in getting people back for follow-up doses after the first one.
13. Poor integration of immunization programs into other health activities.
14. Lack of political will and support.
15. Higher priority given to other health or economic programs.

mortality, the control of the infection itself, and the control of the presence of the causative organism in the environment.[69] True eradication is achieved only when there is no risk of infection or disease in the absence of vaccination or any other control measure in the entire world. The disappearance of transmission in a given area is termed "elimination" but would not exclude the importation of infection from outside.

The biological features favoring the possibility of eradication or a high degree of control are listed in Table 15. The most important are the limitation of the infection to the human host, the absence or sparsity of subclinical infections, and only one serotype of the virus. Smallpox fulfills all these criteria and is the "gold standard" against which others are judged. In addition to those listed is the need for the vaccine to be inexpensive enough to be used worldwide, to be thermostable, to be capable of inducing immunity in a few injections because of the failure of some 50% of persons in developing areas to return for serial injections, and that it is simple enough to administer in a primitive setting.[35]

There are also socioeconomic factors favoring control: the impact of the disease on the economy must be significant enough to motivate political action in its control, the control programs must fit in with other activities in preventive and curative medicine, and they must be compatible with other economic priorities such as the provision of an adequate food supply, control of the birth rate and infant mortality, and other housing and defense needs. The infections, other than smallpox, that most closely fit these criteria are measles, poliomyelitis, rubella, and yaws in about that order.

Great progress has been made in several developed countries in the "elimination" or near elimination of mea-

Table 15. Factors Favoring Eradication of Communicable Diseases[a]

Infection and disease limited to human host and transmitted person to person (no animal or insect reservoir).
Characteristic clinical disease, usually serious, and easily diagnosed.
Few or no subclinical cases.
No long-term carrier states.
Only one causative agent or serotype.
Short period of infectivity pre- and postdisease.
Immunity following disease or immunization is:
Of long duration.
Not subject to reinfection or reactivation.
Decreases or eliminates excretion of organism.
Evidence of vaccine immunity detectable.
Disease has seasonality (permitting vaccine strategies).
Characteristics of vaccine needed:
Simulates natural infection.
Stable: resists physical and genetic change.
Eradication would be cost effective.

[a]Reprinted from ref. 69.

sles, but often pockets of susceptibility remain either because the immunization began too late in life or the young adults were not included and were not old enough to have had natural infection or because of refusal on religious or other grounds. In developing countries, however, there are major obstacles, even in addition to those listed in Table 14. Among the most difficult problems are that many cases of measles occur under the age of 1 and that maternal immunity is of much shorter duration, perhaps because the mother was also infected in infancy and the immunity has waned, or possibly there is more rapid loss of antibody. Whatever the reason, measles immunization is needed in that rather brief "window" of time between the loss of maternal antibody and exposure to natural measles infection, and this period may vary in different countries, even in the same setting and possibly from individual to individual. If vaccine is given too early, a poor antibody response may occur in the infant, and booster doses may be relatively ineffective or the immunity short-lived. The use of an intranasal vaccine may circumvent this issue, but there are technical problems in its proper administration; more potent injectable vaccines may overcome low levels of maternal antibody (see Chapter 16 for a fuller discussion).

Poliomyelitis is next on the list, but the presence of many subclinical cases, three serotypes, and relatively long persistence of the virus in the intestine pose challenges in its control. However, given enough personnel, a massive 1- or 2-day countrywide immunization program can be mounted in some areas as in Brazil,[185] and remarkable control can be achieved in a short time. Such an effort simulates a large vaccine-induced epidemic, since the virus spreads to contacts. Whether such a program would overcome the poor seroconversion rate to oral vaccine found in many African countries has not been established, nor can every country afford to place primary emphasis on the control of one disease at the expense of other vaccination and primary care programs. The new inactivated vaccine in one or two doses in settings where the response to oral vaccine has been poor or where it has not been logistically possible to administer three or four doses of oral vaccine should be considered, at least as the first encounter with a vaccine. This can be accompanied or followed by oral vaccine. A full discussion of these issues can be found in Chapter 9.

Rubella is another possible candidate because it is limited to the human host and has one serotype, but about half the infections are subclinical, and disease is not of high economic impact; protection of congenital rubella in the newborn is the major objective. Immunization both in childhood and young adulthood may be necessary to give long enough protection. It seems unlikely that elimination of measles or poliomyelitis can be fully achieved in the developing world, but control of the clinical disease and its associated mortality seems a worthy and attainable objective over time.

15.1.3. Strategies for Vaccine Delivery. Vaccines will only be effective if they are administered to the persons who need them. Various strategies have evolved in developed and in developing countries to achieve this end. They must be adjusted to the social, economic, cultural, religious, climatic, and logistic setting in which they are used. In the United States the various states, operating under the guidance and encouragement of the CDC, have initiated the requirement for completion of vaccines against childhood diseases as a criterion for entry into the school system. The proof required and the vigor of the enforcement of the regulations have been the major determinants of whether success has been achieved within the existing guidelines for preventable diseases. A dramatic drop in measles was achieved by this means by 1982, with a 97% decline in cases over the number in 1977. However, pockets of susceptible persons still exist, particularly among college-age students, accounting for 18% of the cases in the first half of 1985. During this period 1807 cases were reported, of which 25.9% were deemed preventable and 74.1% nonpreventable. In the latter group, 18% were too young for routine vaccination (under 16 months of age), and 3.1% were too old (born before 1957); 80.1% or 842 cases had been "adequately vaccinated" on or after their first birthday, and these are considered to be "nonpreventable" under current guidelines.[36] Requirement for proof of measles vaccination prior to entry into college is underway.

Czechoslovakia has achieved almost total elimination of measles through required immunization, and Canada and other developed countries are embarked on similar pro-

grams. Elimination of rubella from developed countries is also a feasible objective, and in the United States only 954 cases were reported in 1983.[34] However, the requirement on entrance into grammar school may have to be reinforced with a booster dose on entrance to high school, since 10–15% of this age group is still susceptible in the United States.[34]

Routine immunization has essentially eliminated paralytic poliomyelitis in the United States with the exception of a few vaccine-associated cases and occasional paralysis in an immunocompromised recipient. In developing countries a variety of vaccine strategies have been tried in an effort to control the six diseases targeted by the World Health Organization. These include (1) integration of vaccines into the primary care program with special emphasis where necessary, (2) obtaining political endorsement for the vaccine program both at the national level and at the smallest administrative unit where vaccine is to be given, (3) seeking help from volunteers and from the community, (4) creating special vaccine days, a "national vaccine day," or "pulse vaccine days" such as used in some parts of India when intensive periodic programs are carried out in different communities, (5) using mobile teams to go from village to village, (6) vaccinating from household to household, (7) delivery to concentrated population groups and setting up satellite vaccine stations in remote areas, (7) providing extra services such as oral rehydration salts to mothers bringing infants for immunization, and (8) vaccinating children when they are brought in to clinics or hospitals for medical care, because the risks of reactions to the vaccine are usually less than those of leaving the child unvaccinated, (9) establishing free hospital vaccination clinics for such children as well as for tetanus immunization of pregnant mothers, (10) creating a health registry indicating what children need to be vaccinated, their current vaccine status, when the next shot is due, and the child's height and weight, and (11) setting up means to preserve the viability of vaccine by preserving and monitoring the "cold chain."

15.1.4. Assessment of Immunization Programs. Surveillance and seroepidemiogic techniques are important in the planning and evaluation of immunization programs. The groups at highest risk for infection and disease should first be identified as the major target group; often this is the newborn child in developing countries. The percentage who receive the vaccine and any attendant side reactions should be measured as the program is implemented. The criteria for assessing the control of infectious diseases through immunization programs have been reviewed for poliomyelitis[67] as well as other viral diseases,[68] and the need for serological evaluation has been stressed.[66] The evaluative procedures should be applied at three levels: the control of clinical illness and its attendant mortality, if any, the control of the infection itself, and the control of the transmission and presence of the virus in the environment. The surveillance methods for determining the control of the clinical illness are given in Table 16.

As mentioned earlier, WHO has been a leader in practical ways to carry out some of these methods, especially the household survey in which comparable information can be obtained from different countries by utilizing a standard procedure.[216] There are limitations to these various methods that should be recognized in interpreting their significance. They involve both the numerator (the cases) and the denominator (the population at risk). The former category includes the inadequacy of medical facilities for diagnosis, the lack of laboratories for confirmation of the diagnosis, a most important element of most viral diseases, and inadequate reporting, even when the disease has been diagnosed, a problem common in both developed and developing countries. Finally, there may be an inadequate health structure for analysis of the data or an inadequate system of communication with the health providers. Similarly, knowledge of the denominator may be inadequate for reasons relating to the actual census data, the age, sex, and socioeconomic distribution of the populations at risk, even good figures on the birth, death, and infant mortality rates in developing countries. Household surveys may present problems when the residents are not at home, when there are language problems, or when the marker of the disease is imprecise. Often there are memory defects as to past diseases or immunizations.

The assessment of the control of the infection requires more sophisticated techniques, as many infections do not result in clinical illness. The immune status of a population can be judged indirectly by immunization surveys or knowledge of vaccine usage and can be directly measured by seroepidemiologic surveys. Using a household cluster technique employing questionnaire and interview data, WHO has estimated that the percentage of children immunized with three doses of polio vaccine by 12 months of age was 82% in Europe, 24% in the eastern Mediterranean, and 3% in Southeast Asia[216]; comparable data were found for three doses of DPT and one measles vaccination. A major problem has been that one-third to one-half of the children have failed to return

Table 16. Surveillance Methods for Assessing Control of the Clinical Illness

1. Required reporting of morbidity and mortality of the disease.
2. Special or sentinel reporting through representative health care units.
3. Hospital and physician surveys for admission of patients with the disease.
4. Household surveys.
5. School surveys for evidence of the residuum of the disease (paralysis for poliomyelitis).

for a second dose of vaccine. Studies of vaccine usage indicate that most countries are now using vaccines against WHO's six targeted diseases that meet their standards. Maintenance of the cold chain has been a problem that WHO is assessing with continuously recording thermometers to monitor the temperature, and the use of magnesium and sorbitol is helping to improve the stability of live vaccines. The third technique is through the use of properly conducted and representative seroepidemiologic surveys. These provide objective evidence of the need for immunization, of the age groups at highest risk, and of the immunologic response to the vaccines administered. They are discussed in Chapter 2.

The third set of criteria for the control of infection is the assessment of the transmission and the presence of the agent in the environment for those viruses that are found in water, milk, food, or insect vectors. The method of measurement depends on the route of transmission. For poliomyelitis, samples of sewage are tested for wild virus; for yellow fever and other insect-borne diseases, collection pools of the appropriate mosquito or tick are tested for the presence of the agent. In summary, it is important to emphasize that a vigorous surveillance system, backed where possible by laboratory data, is an integral and important part of any immunization program.

15.2. Chemoprophylaxis and Therapy

Rapid progress is being made in the development of antiviral drugs effective against various points in the replicative cycle of viruses, a process that includes adsorption, penetration, uncoating, transcription, translation, genome replication, virion assembly, and maturation.[50,159] Increased knowledge of this replicative cycle and of the sites of action of various drugs has led to the production of many compounds in various stages of development and testing. The steps include *in vitro* experiments, animal testing, human clinical trials, and, finally, if all goes well, licensure. Rapid diagnostic techniques now permit early use of the drugs shown to be effective against a specific virus in a given clinical setting. The limitations to the development of antiviral agents is that *in vitro* and animal models do not always predict their effectiveness in actual human use, that different viruses, even strains of the same virus, may respond differently, that no drug is truly virucidal, and that resistance to the drug may emerge. Toxicity may present a problem because of the difficulty of drugs in distinguishing sufficiently between certain host cell functions and viral replication. Longer-term toxic effects such as oncogenicity and teratogenicity are also of concern. In contrast to bacterial antibiotics, few available antiviral agents are broad spectrum in their activity, but some are useful in both prophylaxis and therapy against a single virus. The major current challenges are to find preparations that would cure AIDS or that would prevent the development of clinical disease among those infected with HIV.

Table 17 shows the most important licensed antiviral preparations and their uses. In addition, idoxuridine and trifluridine are licensed for topical treatment of herpes simplex keratitis, but viral strains resistant to idoxuridine have appeared. Amantadine hydrochloride is effective in prophylaxis, and to a lesser extent in therapy, against most influenza A strains but not against influenza B strains. Rimantadine, a related drug not yet licensed in the United States but in use in Europe, Britain, and the Soviet Union, is said to more effective against influenza A strains and less toxic than amantadine. The precise mechanism of action of these drugs is not entirely clear, but they may interfere with uncoating of the virus after penetration into the cell or with primary transcription of the RNA. When used in prophylaxis they are about 50% effective in preventing infection with the virus and over 60% effective against the development of clinical illness. This difference may be useful, since infection without disease confers immunity.[159] Side reactions to amantidine occur in 5–10% of recipients and consist of mild central nervous system symptoms such as anxiety, insomnia, and difficulty in concentrating. Rimantadine appears to be less toxic and may prove to be useful in elderly patients, especially in nursing homes, in combination with influenza vaccination. A moderate therapeutic effect has been shown for both drugs in reducing the fever and symptoms of clinical influenza, at least in trials in young adults. Currently both the logistics of use and the toxicity of amantadine limit their application to the high-risk, well-defined groups shown in Table 17.

Vidarabine (adenine arabinoside) is active against all the human herpesviruses. It inhibits nucleic acid synthesis through one or more mechanisms. Clinically, it has several uses in herpetic infections (see Table 17). In proved cases of herpes simplex encephalitis (HSE), vidarabine has reduced the mortality from 70 to 28% in placebo-controlled trials at the end of 1 month and to 40% at 6 months.[10] About half of the survivors have relatively normal function at the end of a year. The comparative effectiveness of vidarabine with acyclovir is under study. Limitations to its use are the current need for a brain biopsy to establish the diagnosis and the large amount of infusion required to administer the drug because of its low solubility. This is a special problem in patients with increased intracranial pressure. In newborn infants with central nervous system or disseminated infections with HSV, the mortality has been reduced from 74 to 38%, but only 29% of the survivors are normal at the end of a year. The drug is effective intravenously in herpes zoster infections (shingles) in immunocompromised patients, as demonstrated by beneficial effects on cutaneous and visceral manifestations, by reduction in new lesion formation, and by reduction in the duration of viral shedding as well as by the shortening of

Table 17. Antiviral Agents Approved for Prophylaxis or Therapy

Agent	Viral infection	Use	Application	Toxicity	Effectiveness
Amantadine	Only influenza A	Prophylaxis (oral)	During proved influenza A outbreak for persons not vaccinated against current strain who: (a) Have underlying disease (e.g., cardiac, respiratory) that puts them at risk to serious illness (b) Are older persons in institutional settings (c) Are adults essential for medical care and other inpatient community services (d) Are possibly certain hospitalized patients	5–10% develop CNS symptoms: confusion, hallucinations, anxiety, insomnia; reversible	>60% vs. clinical illness 50% vs. influenza infection
		Therapy	Consider use in first 24–48 hr after onset in persons (a) and (c) above plus those with influenza pneumonia		50% reduction in fever; duration shortened 1–2 days
Vidarabine (Ara-A, Vira-A, adenine arabinoside)	Herpes simplex, herpes zoster	Therapy (topical) (I.V.)	Proved herpes simplex encephalitis (HSV) Infants with proved disseminated HSV infections or CNS Topically for HSV acute keratoconjunctivitis or recurrent epithelial hepatitis Herpes zoster in immunocompromised patients	Low: occasional nausea, vomiting, disorientation, or skin rash; rare CNS symptoms	Reduced HSV encephalitis mortality from 70 to 28%
Acylovir	HSV	Therapy (I.V. and topical	Mucocutaneous HSV in immunocompromised pts., primary genital herpes infections	Low	Good
Ribavirin	RSV	Aerosol	Severe lower respiratory infections in infants and children	Do not use with assisted ventilation or in or prior to pregnancy	Good

postherpetic neuralgia. It has also been of benefit in the treatment of varicella infections in immunocompromised patients. The Food and Drug Administration of the United States has approved its usage in these VZV infections in immunocompromised patients.

Acyclovir is a drug recently licensed in the United States. It is a potent and specific inhibitor of certain herpesviruses in which a virus-coded thymidine kinase, present in infected tissues, phosphorylates the drug to its active form, acyclovir monophosphate. It is active against HSV-1,

HSV-2, and VZV, all of which induce deoxypyrimidine kinase, but has little effect against CMV, which does not produce this enzyme. Epstein–Barr virus is more sensitive than CMV to the drug, although it does not induce its own deoxypyrimidine, perhaps through its action on EBV DNA polymerase. Acyclovir is available for intravenous (I.V.) and topical use, and an oral form is under consideration for licensure. Intravenous administration has proved of marked benefit in primary genital infections and in mucocutaneous HSV infections in immunosuppressed patients; topical therapy has been less effective. Virus shedding, healing time, new lesion formation, and the duration of symptoms are reduced under treatment.[50] However, virus shedding and new lesions may develop after discontinuance of the drug. Oral acyclovir therapy is also effective in primary infections but of questionable value on the rate of subsequent recurrences. Early patient-initiated therapy for recurrences may shorten the episode by about 30%. Promising results are also being found in preliminary trials of long-term prophylaxis.

Ribavirin is a purine nucleotide analogue that has shown a wide spectrum of activity *in vitro* against both RNA and DNA viruses. Its precise mechanism of action is not clear. Its aerosol use has been approved by the U.S. Food and Drug Administration for carefully selected cases of severe lower RSV infections of infants and young children. Placebo-controlled trials have shown significantly greater improvement than controls in the severity of illness, in arterial oxygen saturation, and in a shorter duration of virus shedding. No significant toxicity was noted during therapy. Careful respiratory monitoring should be maintained throughout treatment. Ribavirin aerosol should not be used for infants requiring assisted ventilation because precipitation of the drug in the respiratory equipment may interfere with safe and effective ventilation of the patient. Deterioration of respiratory function has been associated with ribavirin use in infants and to some extent in adults with chronic obstructive lung disease or asthma. Ribavirin is not indicated in milder respiratory infections caused by RSV, which constitute the majority of cases and in which the clinical course runs less than the 3 to 7 days required for a full course of ribavirin therapy. Hematopoietic toxicity has been noted in oral administration. Conflicting results of oral therapy in influenza A and B infections have been obtained, and currently use in only severe RSV infections has been approved. Animal, tissue-culture, and some clinical trials have suggested its usefulness in hepatitis A, measles, Lassa fever, and herpes simplex infections, and there is hope that it might be helpful in HIV infections, but more experience with its use in these situations will be needed to evaluate its effectiveness.

In addition to antiviral drugs, the application of interferon to viral prophylaxis and therapy is receiving more attention now that recombinant DNA technology has made greatly increased amounts available for clinical trial and commercial development. The three major forms of interferon are α (from leukocytes), β (from fibroblasts), and γ (from T lymphocytes). They have diverse biological effects in addition to antiviral activity. The antiviral action is apparently mediated through degradation of messenger RNA at the translational level of viruses. The new recombinant interferon is similar in effect to α interferon. Interferon has been tested in clinical trials for some severe infections using different routes of administration. These include herpetic dendritic keratitis, chronic hepatitis B infections, congenital CMV infections and CMV infections in immunosuppressed patients, rhinovirus and coronavirus common colds, and single assorted infections. No consistent, clear-cut benefit in these circumstances has been established. In prophylaxis it has not prevented recurrences of herpesvirus infections in renal transplant recipients. The most encouraging results have been found in intranasal prophylaxis of rhinovirus infections in susceptible and exposed individuals in families with an index case. The availability of this compound will now allow reevaluation of its clinical efficacy in different infections, especially for prophylaxis.

Additional information on the control and prevention of specific viral infections is included in the appropriate sections of subsequent chapters of this book.

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