

Food-borne Viruses: Prevention and Control

Efstathia Papafragkou, Doris H. D'Souza, and
Lee-Ann Jaykus

1.0. INTRODUCTION

Epidemic and sporadic gastroenteritis is an important public health problem in both developed and developing countries. In the United States, as many as 67% of all food-borne illnesses, 33% of the associated hospitalizations, and 7% of deaths attributable to food-borne disease may be caused by viruses, resulting in approximately 30.9 million cases each year (Mead et al., 1999). Costs of illness are high simply by virtue of the frequent occurrence and high transmissibility of enteric viruses (Koopmans et al., 2002). The total burden of enteric viral disease can only be estimated as most of the illnesses are mild, go unreported, and routine testing of patients for specific virus infections is not usually done (Richards, 2001). Year-round outbreaks have affected adults and children in various settings. These viruses circulate readily in families, communities, and in places where individuals are in close proximity or are using a common source of food or water. Consequently, many documented outbreaks of enteric viral illness have occurred in schools, recreational camps, hotels, hospitals, orphanages, and nursing homes and among individuals consuming a common food item served in a restaurant or banquet setting. Hepatitis A virus (HAV), human caliciviruses, and group A rotaviruses are among the most important food-borne enteric viruses.

Unlike bacteria, viruses cannot replicate in food or water. As a result, when virus contamination of food occurs, the number of infectious virions will not increase during processing and storage. The ability of contaminated food to serve as a vehicle of infection, therefore, depends on virus stability, degree of initial contamination, the method of food processing and storage, viral dose needed to produce infection, and the susceptibility of the host (Koopmans et al., 2002). Virus particles are stable to environmental extremes including pH, low temperatures, and some enzymes, particularly those found in the human gastrointestinal tract (Jaykus, 2000a). Consequently, human enteric viruses can withstand a wide variety of food storage and processing conditions making virtually any kind of food product a potential vehicle for virus transmission (Jaykus, 2000b). Because food-borne viruses are transmitted via the fecal-oral route through contact with human feces and because infected individuals can shed millions of virus particles in their stools, the role of infected food-handlers cannot be underestimated.

From an epidemiological perspective, human caliciviruses are the most significant by virtue of the sheer number of cases caused by this virus group. Caliciviruses are composed of multiple genera; however, the noroviruses and the saporoviruses are the primary cause of human disease and are responsible for up to 2.3 million infections, 50,000 hospitalizations, and 300 deaths per year in the United States alone (Mead et al., 1999). Of all the gastroenteritis outbreaks reported in England and Wales, nearly 50% were due to noroviruses, a figure that is similar to those reported for other European countries including Finland, Sweden, The Netherlands, and Germany (Lopman et al., 2002). Indeed, noroviruses are the most common cause of acute, nonbacterial gastroenteritis worldwide. Although the disease caused by this virus group is short-lived and recovery is usually complete, immunity is poorly understood and there is significant antigenic and genetic diversity within the genus. As a result, some individuals may remain susceptible and/or become infected multiple times during the course of their lives (Jaykus, 2000b).

Noroviruses are transmitted directly by person-to-person contact or indirectly via contaminated food, water, or fomites. Person-to-person transmission can occur by two routes: fecal-oral and aerosol formation after projectile vomiting (Patterson et al., 1997). A total of 348 outbreaks of norovirus gastroenteritis were reported to the U.S. Centers for Disease Control and Prevention (CDC) between January 1996 and November 2000. Of these, food was implicated in 39%, person-to-person contact in 12%, and water in 3% of the outbreaks. Interestingly, 18% of the outbreaks could not be linked to a specific transmission mode (Centers for Disease Control, 2001). Frequently during an outbreak, secondary cases occur as a consequence of contact with the primary cases (Koopmans and Duizer, 2004). A low infectious dose (10–100 virus particles; Koopmans et al., 2002) and a propensity for long-term virus excretion (up to 2 weeks postinfection) are the likely reasons for the high attack rates (~50%) seen in norovirus outbreaks (Koopmans et al., 2002). Although attention has recently focused on high-profile cruise ship outbreaks (Centers for Disease Control, 2002), an estimated 60–89% of all acute viral gastroenteritis outbreaks occur on land (Centers for Disease Control, 2003b). Although norovirus infection predominates during cold-weather months (Mounts et al., 2001), hence the term “winter vomiting disease,” it has been diagnosed year-round (Bresee et al., 2002).

Hepatitis A virus (HAV) is another enteric virus that can be transmitted by contaminated food. More than 95% of HAV infections are transmitted by the fecal-oral route (Ciocca, 2000) and person-to-person contact is considered to be the primary mode of transmission (Koopmans et al., 2002). Therefore, outbreaks of HAV are common in high population density settings such as schools, prisons, and military bases (Koopmans et al., 2002). Approximately 50% of the reported HAV cases do not have a recognized source of infection, and only 5% have a clear food- or waterborne route. In the United States alone, HAV causes an estimated 83,000 illnesses per year (Mead et al., 1999). Improved sanitary conditions have caused a decline in the

prevalence of this disease in the developed world, but increases in international travel and trade have brought a naïve population in contact with endemic disease, resulting in a reemergence of HAV infection in developed countries (Romalde et al., 2001). For example, a recent, large outbreak of HAV was attributable to imported green onions. This incident alone resulted in more than 600 infections and three deaths (Centers for Disease Control, 2003c).

Rotavirus is the most common cause of severe infant diarrhea (Lopman et al., 2002), estimated to be responsible for 130 million illnesses and more than 600,000 deaths per year throughout the world (Mead et al., 1999). Rotaviruses are most often transmitted by the waterborne route but have occasionally been implicated in outbreaks attributed to contaminated food (Bresee et al., 2002). It is estimated that only 1% of rotavirus cases are food-borne (Sair et al., 2002). Spread of the disease is mainly through the fecal-oral route, although aerosol transmission has also been suggested (Caul, 1994).

In reviewing food-borne transmission of human enteric viruses, three major at-risk food categories stand out: (i) shellfish contaminated by fecally impacted growing waters, (ii) human sewage pollution of drinking and irrigation water, and (iii) ready-to-eat (RTE) and prepared foods contaminated by infected food-handlers as a result of poor personal hygiene (Jaykus, 2000b). Among the more common foods that have been implicated as vehicles for enteric virus transmission are shellfish, fruits, vegetables, salads, sandwiches, and bakery items. Indeed, any food that has been handled manually and is not further heated prior to consumption has the potential to be virally contaminated (Richards, 2001). In further discussion of prevention and control strategies for enteric viruses, we categorize the discussion into these three distinct commodity groups because prevention and control differ for each.

2.0. SHELLFISH

Bivalve mollusks (including mussels, clams, cockles, and oysters) have been implicated as vectors in the transmission of bacterial and viral enteric diseases for many decades. The most commonly implicated bivalves are oysters, followed by clams (Potasman et al., 2002). It is estimated that human enteric viruses are actually the most common human disease agents transmitted by molluskan shellfish (Lees, 2000; Formiga-Cruz et al., 2002). Although more than 100 different types of enteric viruses can be excreted in human feces, only a few (HAV, noroviruses, astroviruses) have been epidemiologically linked to shellfish-associated viral disease (Richards, 2001). Of these, infectious hepatitis caused by HAV is probably the most serious. Immunosuppressed patients are at high risk for serious disease and as a precaution should be advised to avoid this particular cuisine (Potasman et al., 2002). In fact, the largest viral food-borne disease outbreak occurred in Shanghai,

China, in 1988, where 300,000 people were infected with HAV after consumption of clams harvested from fecally impacted growing waters (Halliday et al., 1991). In the 1990s, noroviruses were implicated as the primary etiological agents among reported cases of infectious diseases associated with shellfish consumption (Centers for Disease Control, 2001). Shellfish contamination with noroviruses is of great significance to food safety not only for its direct implications but also for the secondary cases that readily occur after a primary food-borne outbreak (Beuret et al., 2003). Significant and recent outbreaks of viral food-borne diseases associated with shellfish consumption are summarized in Table 13.1.

Unlike illnesses caused by naturally occurring *Vibrio* spp., enteric viral illnesses originate from human fecal wastes only. Even though most sewage treatment processes cannot completely eliminate viruses, adequate sewage treatment remains the first line of defense in protecting shellfish and their harvesting waters (Sorber, 1983). Factors contributing to human sewage pollution of marine waters include the illegal dumping of human waste directly into shellfish harvesting areas, failing septic systems along shorelines, sewage treatment plants overloaded with storm water, and discharges of treated and untreated municipal wastewater and sludge (Jaykus et al., 1994; Shieh et al., 2000). For instance, a recent outbreak in France and Italy was associated with norovirus-contaminated oysters harvested from a pond that was polluted from an overflowing water purification plant (Doyle et al., 2004).

Shellfish are at particular risk of transmitting human enteric viruses because (i) they are frequently eaten whole and raw or only lightly cooked; (ii) there are no good methods to ascertain whether the shellfish or their harvest waters contain infectious viruses; and (iii) shellfish can bioconcentrate viruses within their edible tissues to levels much higher than in the water itself (Richards, 2001). This bioaccumulation is probably assisted by the ionic binding of virus particles to mucopolysaccharide moieties of the shellfish mucus (DiGirolamo et al., 1977). The degree of virus uptake and survival in shellfish depends on several factors, including exposure time, virus concentration in overlay water, presence of particulate matter (and/or excess turbidity), temperature, interspecies differences, individual shellfish differences, type of virus, food availability, and pH and salinity of water (Sobsey and Jaykus, 1991; Jaykus, 1994). Because uptake of virus by shellfish is dependent on active feeding, any factor affecting the physiological activity of the animal can influence virus accumulation. Likewise, the elimination kinetics of enteric viruses by bivalve mollusks can vary with the type of shellfish, type of microorganism, and environmental conditions and season (Burkhardt and Calci, 2000; Mounts et al., 2001).

Preharvest contamination is the most common source of contamination and occurs in shellfish exposed to human fecal pollution of waters from which they are harvested. Once contaminated, the viruses can persist in both the overlay waters and the shellfish. Enriquez et al. (1992) reported rapid uptake (in less than 24 hr) of HAV in the mussel *Mytilus chilensis*, with virus persistence for about 7 days. The ability of HAV to persist in Eastern oysters

Table 13.1 Epidemiological studies summarizing recent enteric virus outbreaks associated with molluscan shellfish

<i>Agent</i>	<i>Food</i>	<i>Samples tested</i>	<i>Detection Methods</i>	<i>Conclusions</i>	<i>Reference</i>
Norovirus	Oysters	Clinical samples	Electron Microscopy (EM), RT (reverse transcription)-PCR, Sequencing	Overboard disposal of sewage from a harvesting boat into the oyster beds	Aristeguieta et al., 1995
Norovirus	Oysters	Outbreak (clinical and food)	IgG antibodies, EM, RT-PCR, Sequencing	Contamination in the oyster beds from overboard disposal of sewage from handlers	Berg et al., 2000
Norovirus	Oysters	Clinical samples and oyster samples	Bacteriological sampling, Screen for F+ and somatic phages	Virus particles can persist in the oysters for many weeks after depuration	Chalmers and Memillan, 1995
Norovirus, HAV	Oysters	Clinical samples	RT-PCR, Sequencing	Imported contaminated clams, not properly steamed	Furuta et al., 2003; Kohn et al., 1995
HAV	Clams	Clinical samples	Enzyme Immunoassay (ELISA), EM	Untreated sewage from fishing vessels and the surrounding residential area	Halliday et al., 1991
HAV	Clams	Outbreak (clinical)	ELISA	Secondary infection through person to person contagion	Leoni et al., 1998

(*Crassostrea virginica*) was recently investigated by Kingsley and Richards (2003), who detected virus in the animal as little as 16 hr after exposure, with the oysters remaining infectious for up to 3 weeks thereafter. Of the factors that influence the survival and persistence of enteric virus in seawater, temperature is particularly important (Muniain-Mujika et al., 2003). For instance, it has been demonstrated that the time necessary to inactivate 90% of HAV in seawater was 671 days at 4°C but only 25 days at 25°C (Gantzer et al., 1998). Similarly, poliovirus type 1 at a concentration of 10^5 pfu/ml lost its infectivity in ocean water within 27 days during the summer but within 65 days during the winter (Lo et al., 1976). Recent research has indicated that about 10% (17/191) of Japanese oysters sampled and intended for raw consumption harbored noroviruses, and in some of these samples, the virus level was quite high (Nishida et al., 2003).

2.1. Preharvest Control Strategies

2.1.1. Harvest Water Quality: Preventing Illegal Sewage Discharge

The most effective and reliable approach to control viral contamination of shellfish is to harvest them from areas with good water quality, most notably from estuarine environments that are free of human sewage contamination. Unfortunately, recent virus outbreaks have been caused by the dumping of untreated human sewage from boats, resulting in contamination of shellfish beds. Enforcement of proper waste disposal in certain discharge locations may be difficult as harvesting areas are frequently remote from the shore and there may be many harvesting and recreational vehicles present at any one time. A preemptive measure would be to provide dockside receptacles for waste disposal. Alternatively, mandating the use of a waste container that cannot be easily dumped or flushed into the harvesting waters could protect water quality (Berg et al., 2000). Furthermore, imposing severe monetary penalties for violation of overboard waste dumping and improper onboard waste receptacles may also be an effective deterrent. Lastly, educating harvesters about the public health risks associated with overboard sewage disposal, as well as the need for compliance with regulations for waste disposal, are integral steps to preharvest control.

2.1.2. Harvest Water Quality: Microbiological Indicators

Historically, the fecal coliform index has been employed as an indicator of the sanitary quality of shellfish and their harvesting waters. This index has been considered appropriate as fecal coliforms are normal inhabitants of the gastrointestinal tract of warm-blooded animals and are excreted in the feces in large numbers (Jaykus, 1994). Standards exist in the United States for shellfish and shellfish-harvesting waters based on the enumeration of total and fecal coliforms. The National Shellfish Sanitation Program (NSSP), a federal/state cooperative program recognized by the U.S. Food and Drug Administration (FDA) and the Interstate Shellfish Sanitation Conference (ISSC), is responsible for the promotion of sanitary quality of shellfish sold for human consumption. NSSP sponsors numerous programs to promote

shellfish safety, including evaluation of state program elements, dealer certification, and state growing area classification, including sanitary surveys of shellfish-growing waters (National Shellfish Sanitation Program, 2000). Accordingly, a fecal coliform standard of less than 14 MPN (most probable number) per 100 ml of water, with not more than 10% of samples exceeding 43–49 MPN/100 ml (depending on the microbiological method), is required for classification of harvesting waters as approved. Outside of these standards, waters can be classified as conditionally approved, restricted, or prohibited for shellfish harvesting (Somerset, 1991). Current regulations also stipulate fecal coliform counts of less than 45 MPN per 100 g of shellfish meat for fresh product to be commercially marketed.

The fecal coliform standards for shellfish, although effective in controlling bacterial disease transmission, do not necessarily prevent virally contaminated shellfish from reaching the marketplace (Kingsley et al., 2002a). In fact, these standards may offer no indication of viral contamination, as viruses can persist for a month or longer within shellfish or estuarine sediments, long after coliform counts have reached acceptable levels (Kingsley and Richards, 2001). As a result, there is no clear and consistent relationship between the occurrence of bacteriological indicators and viruses in water or shellfish (Sobsey and Jaykus, 1991; Jaykus et al., 1994). Romalde et al. (2002) examined European shellfish contaminated with HAV and human enteroviruses and confirmed that there was no correlation between the presence of the traditional bacterial indicators and enteric viruses. Power and Collins (1989) arrived at similar conclusions when investigating depuration of poliovirus, *Escherichia coli*, and a coliphage by the common mussel, *Mytilus edulis*. Apparently, *E. coli* was not a good indicator of the efficiency of virus reduction during depuration, while the coliphage appeared to be a more reliable one. These data and others confirm that compliance with the fecal coliform end-product standards does not provide a guarantee of the absence of enteric viruses, even in depurated shellfish. In fact, in areas where the current European microbiological standards characterizing waters as suitable for harvesting shellfish were in place, the infectivity of HAV in mussels after depuration was recorded to be reduced by only 98.7% (Abad et al., 1997b). Another study done with mussels harvested in Italy revealed that although the product's microbiological quality was in accordance with the European bacteriological standards, HAV was detected in 13 of 36 specimens (Crocchi et al., 2000).

As bacterial indicators generally fail to signal the potential for viral contamination, bacteriophages, enteroviruses, and adenoviruses have all been proposed as alternative indicators (Lees, 2000; Muniain-Mujika et al., 2002). The feasibility of using human adenoviruses as indicators of human enteric viruses in environmental and shellfish samples was suggested by Pina et al. (1998) who reported that these viruses were easily detected and seemed to be more abundant and stable in environmental samples. In fact, the presence of human adenoviruses appears to correlate with the presence of other human viruses, and they have been proposed to monitor viral contamination

in shellfish harvested from Greece, Spain, Sweden, and the United Kingdom (Formiga-Cruz et al., 2002). On the contrary, no solid correlation could be demonstrated between the occurrence of human enteroviruses and noroviruses in estuarine waters in Switzerland (Beuret et al., 2003).

Bacteriophages, especially F-specific coliphages, somatic coliphages, and phages of *Bacteroides fragilis*, have long been proposed as possible indicators of viral contamination of the environment. Of these, the distribution and survival of F-specific coliphages (also known as FRNA phage or male-specific coliphage) are considered to be more similar to human enteric viruses in the environment. The survivability of FRNA phage in seawater was found to be similar to that of a variety of enteric viruses, including HAV, poliovirus, and rotavirus (Chung and Sobsey, 1993). A study examining the distribution of FRNA phages in shellfish harvesting areas revealed that these phages were more resistant to environmental stresses and that their numbers in shellfish were consistently higher than those of *E. coli*, indicating that their presence in shellfish may be a more representative assessment of virus contamination (Dore et al., 2003). In another study, Sinton et al. (2002) provided evidence that FRNA phages were more resistant to sunlight inactivation than were fecal coliforms, *E. coli*, enterococci, and somatic coliphages, making them potentially more useful for monitoring the virological quality of freshwaters. In a recent study examining the comparative survival of feline calicivirus (FCV, a norovirus surrogate), *E. coli* and FRNA phage in dechlorinated water at 4°C, 25°C, and 37°C, a correlation was found between the survival of the phage and FCV, indicating that the phage may be a potential environmental surrogate for noroviruses (Allwood et al., 2003).

FRNA phages have also been suggested as a complementary parameter for evaluating the efficiency of depuration in heavily contaminated mussels. In a study by Muniain-Mujika et al. (2002), a 5-day depuration period was considered an adequate decontamination treatment because neither human enteric viruses nor FRNA phages could be detected in the shellfish after that time. The applicability of F-specific coliphage as an indicator of depuration efficacy was confirmed by Dore and Lees (1995), who monitored the elimination patterns of *E. coli* and FRNA phage in contaminated mussels and oysters. After a 48-hr depuration period, it was found that *E. coli* was completely eliminated but FRNA phages were still largely retained in the digestive gland. The F-specific RNA bacteriophage have also been suggested as an alternative indicator of potential norovirus contamination in depurated, market-ready oysters (Dore et al., 2000). Formiga-Cruz et al. (2003) studied the correlation between FRNA phages, somatic coliphages, and bacteriophages of *B. fragilis* as indicators of viral contamination in shellfish and found that FRNA phages were better predictors of norovirus contamination than for adenovirus, enterovirus, or HAV contamination.

Evidence that phages infecting *B. fragilis* RYC2056 may be a suitable group of indicators for viral pollution in shellfish has recently been provided; however, further research is needed to develop the appropriate methodology (Muniain-Mujika et al., 2003). Earlier research from the same group

suggested that phages infecting *B. fragilis* RYC2056 are preferable to phages infecting *B. fragilis* HSP40 as potential indicators of viral contamination in shellfish, as the former are more abundant in shellfish as well as in sewage (Muniain-Mujika et al., 2000).

Callahan et al. (1995) compared the inactivation rates of HAV, poliovirus 1 (PV-1), F-specific coliphage, and somatic *Salmonella* bacteriophages (SS phages) in seawater and found that SS phages survived significantly longer at 20°C (Callahan et al., 1995). Although the final concentration of all four viruses was reduced to similar levels at the end of the study, the rates of inactivation were different. For instance, it took 10 and 4 weeks to achieve a 4log₁₀ reduction of SS phages and HAV, respectively, whereas PV1 and FRNA phages were reduced to the same extent within 1 week. The study concluded that because SS phage persisted longer in seawater environments, it may be a more reliable indicator of enteric virus contamination.

Somatic coliphages have also been suggested as reliable indicators of the efficiency of shellfish depuration as they can be easily and rapidly assayed (Power and Collins, 1989; Muniain-Mujika et al., 2002). However, Legnani et al. (1998) observed no significant differences between the occurrence of somatic coliphages and fecal indicator bacteria in seawater. In another study, phages of *B. fragilis* and *Salmonella* were found not to be adequate to indicate fecal contamination (Chung et al., 1998), whereas FRNA phage and *Clostridium perfringens* spores were more reliable indicators of human enteric viruses in oysters and the best predictors of fecal contamination in water and oysters. In conclusion, the efficacy of the various bacteriophages as indicators of enteric viruses is still under investigation.

2.1.3. Depuration and Relaying

Two preharvest control strategies rely on extending the natural filter-feeding process of the animal in clean seawater in order to purge out microbial contaminants (Lees, 2000). Both methods are based on the ability of the shellfish to eliminate contaminating microorganisms from their digestive tracts through normal feeding, digestion, and excretion activities. Once in clean waters, shellfish can purge at least some of their contaminants, provided that the water and feeding conditions (primarily temperature, salinity, and dissolved oxygen) are favorable (Richards, 2001).

Depuration, or controlled purification, is the process of reducing the levels of bacteria and viruses in contaminated, live shellfish by placing them in a controlled water environment. In order to produce a safe and wholesome depurated product, specific growing area classification, process approval, and process controls are required (Somerset, 1991). Depuration usually takes place in tanks provided with a supply of clean, often disinfected, seawater under specific operating conditions (Sobsey and Jaykus, 1991). The more common methods of water disinfection for use in shellfish depuration are ultraviolet light, chlorine, or ozonation (De Leon and Gerba, 1990). Ozone, although not a new technology, has generated renewed interest for use in molluscan, shellfish depuration systems (Garrett et al., 1997).

Because the environmental conditions during depuration are tightly controlled, the process usually takes only 2–3 days (Jaykus et al., 1994; Richards, 2001). The process does not suffer from possible recontamination due to changing environmental conditions but is not recommended for shellfish harvested from heavily contaminated (prohibited) waters (Roderick and Schneider, 1994).

Relaying, or natural purification, refers to the transfer of shellfish from contaminated growing areas to approved areas (Sobsey and Jaykus, 1991). Although relaying has lower initial costs compared with depuration, its drawbacks include a lower yield of marketable product and a less steady supply due to environmental variations (Blogoslawski, 1991). In addition, relaying requires extended periods (often 10 days to 2 weeks or more) (Richards, 2001) and is sometimes limited by the availability of suitable pristine coastal areas (Lees, 2000).

Depuration is used extensively around the world and has been successful in reducing bacterial illnesses associated with shellfish consumption in the United Kingdom (Sobsey and Jaykus, 1991). Generally, nonindigenous (enteric) bacteria are rapidly (usually within 48 hr) reduced to nondetectable levels by depuration, while viruses are purged more slowly and may persist for several days (Richards, 2001). It is important to note that the efficiency of both depuration and relaying processes is influenced by numerous factors such as type of shellfish, individual variation in feeding rates, initial level of virus contamination, temperature, turbidity, availability of particulate matter, salinity, pH and oxygen availability (Cook and Ellender, 1986; Sobsey and Jaykus, 1991).

Nevertheless, depurated shellfish can also cause enteric viral illness either as a result of inadequate process control or due to insufficiency of the process itself. Consequently, only lightly contaminated shellfish should be subjected to depuration, whereas the more heavily contaminated ones should be relayed for extended periods of time (Richards, 2001). Most food-borne outbreaks associated with depurated shellfish have been caused by HAV, as this virus does not appear to be as readily eliminated during depuration as do other virus types (Richards, 2001). In an Italian study of 290 mussels collected from various sources, HAV RNA was detected in 20% (20/100) of the nondepurated mussels, 11.1% (10/90) of the depurated samples, and 23% (23/100) of the mussels sampled from different seafood markets (Chironna et al., 2002). These authors concluded that this high prevalence of contamination could be due to the practice of keeping shellfish alive for prolonged periods in possibly contaminated waters or else due to inefficient and/or inadequate depuration. Another Italian study examined the effectiveness of depuration on the decontamination of mussels contaminated with HAV (De Medici et al., 2001). By using a closed-circuit depuration system with constant levels of salinity and temperature and with both ozone and UV disinfection of water, the initial levels of HAV were reduced significantly after 48 hr, but extending depuration to 120 hr allowed for detectable virus reconcentration.

2.2. Postharvest Control Strategies

2.2.1. Temperature

Temperature control, particularly the use of refrigeration temperatures, is a long-accepted method to control the growth of bacterial spoilage microorganisms and pathogens in food. Unfortunately, because enteric viruses do not grow in foods and are in fact quite persistent in low-temperature environments, this approach is not very effective for their control. Indeed, the common practice of icing and freezing are likely to facilitate the survival of viruses, as these are widely used as laboratory preservation techniques (Lees, 2000). With respect to temperature control, a study done with Olympic oysters contaminated with poliovirus showed that even after 15 days of storage at 5°C, the virus titer was reduced by only 60%, while after extended storage for 30 days at 5°C, 13% of the input virus remained infectious (DiGirolamo et al., 1970). The same group of researchers studied the survival of poliovirus in Pacific oysters kept at -17.5°C and concluded that after 4 weeks of storage, the virus titer was reduced by little more than 0.5 log₁₀, while further storage for 12 weeks resulted in a 1 log₁₀ reduction (DiGirolamo et al., 1970). Tierney et al. (1982) reported survival of infectious poliovirus after a 28-day period of storage at 5°C (Tierney et al., 1982). Greening et al. (2001) found poliovirus type 2 to persist in green-lipped mussels, *Perna canaliculus*, even after 2 days of refrigeration (81% of the original titer was recovered), as well as after 28 days of storage at -20°C (44% of the initial titer was detected) (Greening et al., 2001). When T4 coliphage was used as a surrogate for enterovirus contamination in West Coast crabs (*Cancer magister* and *C. antennarius*), less than 1 log₁₀ reduction in virus titer was obtained when the crabs were kept for 120 hr at 8°C, and about 25% of the input coliphage could still be recovered when the crabs were stored at -20°C for 30 days (DiGirolamo and Daley, 1973).

Early thermal inactivation study focused on HAV in steamed clams showed that it took 4 to 6 min of steaming for complete virus elimination, at which point the internal temperature of the clam tissue was 100°C (Koff and Sear, 1967). The authors suggested that it would not be safe to consume steamed clams when they first open, as opening of the shells usually happens within the first minute of steaming. In mussels contaminated with HAV and subjected to steaming for 5 min, 0.14% of the initial HAV could still be recovered (Abad et al., 1997). Pacific oysters artificially contaminated with poliovirus were heat processed by stewing, frying, baking, and steaming, with virus inactivation barely exceeding 90% after conventional cooking times for each treatment (DiGirolamo et al., 1970). Studies in artificially contaminated cockles revealed that HAV was only partially reduced when the shellfish were immersed for 1 min in water at 85°C, 90°C, or 95°C or when steamed for the same period. For complete inactivation of HAV, the internal temperature of the shellfish had to reach 85–90°C and be maintained there for 1 min (Millard et al., 1987). Similarly, another study reported that heat treatment at 100°C for 2 min (internal temperature of 90°C) was needed to assure com-

plete inactivation of HAV in artificially contaminated mussels (Croci et al., 1999).

The failure of several cooking methods (grilling, stewing, and frying) to prevent a large oyster-associated gastroenteritis outbreak was reported in Florida in January 1995 (McDonnell et al., 1997). However, experimental data from the inactivation of feline calicivirus (FCV), a norovirus surrogate, showed that the previous heat-processing recommendations (internal temperature of 90°C for 1.5 min) for the elimination of HAV in cockles could also successfully eliminate FCV in shellfish (Slomka and Appleton, 1998). The survival of FCV was studied at 56°C, 70°C, and 100°C and it was found that 7.5 log₁₀ titer of FCV could be completely inactivated (i.e., nondetectable by infectivity assay) by heating for 5 min at 70°C or for 1 min at 100°C (Doultree et al., 1999). Apart from this study, there has been little work done to determine the thermal inactivation kinetics of the noroviruses. Although not studied *per se*, a 1988 outbreak of HAV linked to a fast-food restaurant in Tennessee suggested that microwave heating may partially inactivate the virus, as customers who reheated their sandwiches before consumption did not develop clinical illness (Mishu et al., 1990).

Not unlike bacteria, the type of virus and the matrix in which the viruses are suspended play a significant role in virus sensitivity to heat (Millard et al., 1987; Croci et al., 1999). For instance, a longer heat treatment was necessary to inactivate HAV suspended in shellfish homogenate compared with the same amount of virus suspended in buffer (Croci et al., 1999). Because shellfish tissue is dense and the virus is likely to be concentrated in the digestive diverticula, heat penetration of this product is of particular concern. It must also be recognized that variability of shellfish species, size, time after harvest, contamination level, and cooking conditions account for the difficulty in establishing a minimum cooking time for complete virus inactivation (De Leon and Gerba, 1990). Moreover, standardization of conditions of commercial heat treatment of shellfish may be difficult because excessive heating may result in undesirable organoleptic changes such as toughening of meat texture.

2.2.2. Ionizing Radiation

Although not a promising technology, ionizing radiation has also been tested as a means to inactivate viruses in shellfish. Oysters (*Crassostrea virginica*), hard-shelled clams (*Mercenaria mercenaria*), and soft-shelled clams (*Mya arenaria*) were contaminated with HAV and rotavirus strain SA-11 and treated with irradiation doses ranging from 1 to 7 kGy. Although a 3-kGy dose resulted in a 95% reduction in virus load, the organoleptic properties of the shellfish also deteriorated at this dose. Using a lower dose of 2 kGy resulted in less than 95% virus inactivation and a product with adequate sensory quality (Mallet et al., 1991). The authors suggested that combining depuration with radiation doses of 2 kGy may effectively decontaminate shellfish, although it is recognized that such an approach would likely be very costly.

2.2.3. High Hydrostatic Pressure Processing

Recently, alternative technologies, particularly high hydrostatic pressure (HPP), have been proposed for the inactivation of HAV and noroviruses in shellfish (Kingsley et al., 2002b). The shellfish industry is very interested in HPP as it has previously been shown to eliminate *Vibrio* species in oysters while maintaining the organoleptic properties of the raw shellfish meat (Berlin et al., 1999). In model studies, HAV and FCV (a norovirus surrogate) suspended in tissue culture medium were eliminated after exposure to 450 MPa for 5 min and 275 MPa for 5 min, respectively. However, model studies with poliovirus showed a general failure of high pressure to inactivate the virus, even at pressures as high as 600 MPa for 15 min (Wilkinson et al., 2001). Extending the pressure treatment for 1 hr had no significant effect on reducing virus infectivity. The authors proposed that perhaps the pressure resistance of poliovirus is correlated with capsid composition. It is clear that further research is needed before HPP can be considered as a viable option for the inactivation of viruses in raw molluscan shellfish.

3.0. PRODUCE

A number of viral food-borne disease outbreaks associated with the consumption of contaminated raw produce have occurred over the past several years, presumably due to the combined effect of increased consumption and better epidemiological surveillance (Centers for Disease Control, 2003c). For example, between 1988 and 1997, the U.S. CDC reported 130 food-borne outbreaks linked to the consumption of fresh produce. A report published by the Public Health Laboratory Service (PHLS) in England and Wales indicated that 83 outbreaks between 1992 and 1999 were associated with the consumption of contaminated salad vegetables or fruit (O'Brien et al., 2000). Of the viral agents, HAV and noroviruses are most commonly documented as contaminating fruits and vegetables. Although fresh produce can certainly serve as a vehicle for the transmission of viral food-borne disease, the exact attribution of this commodity group to the overall burden of this set of diseases is unknown. Furthermore, we know relatively little about the persistence of enteric viruses when they contaminate produce, and the data regarding the efficacy of various virus inactivation methods intended for use on fresh produce are limited and variable (Seymour and Appleton, 2001; Lukasik et al., 2003). Taken together, this means that there is much to learn about viruses in this food commodity.

In most instances, contamination of fruits and vegetables with enteric viruses is believed to occur before the product reaches food service establishments (Koopmans et al., 2002). Sources of such contamination include contaminated soil, contaminated irrigation or washing water, or infected food-handlers who harvest and handle the produce (Lopman et al., 2002). Treatment of sewage sludge by drying, pasteurization, anaerobic digestion, and composting can reduce but not eliminate viruses, especially the more

thermoresistant ones (Metcalf et al., 1995). Therefore, using recycled sewage effluent and sludge to irrigate or fertilize crops intended for human consumption carries with it the risk of virus contamination (Ward et al., 1982). Likewise, soils can also become contaminated by land disposal of sewage sludge and through the use of fecally impacted irrigation water. Viruses can survive in contaminated soil for long periods of time depending on factors such as growing season, soil composition, temperature, rainfall, resident microflora, and virus type (Yates et al., 1985; Seymour and Appleton, 2001).

It is believed that most virus contamination occurs mainly on the surface of fresh produce, although a few studies have reported on the potential for uptake and translocation of virus within damaged plant tissue (Seymour and Appleton, 2001). Use of wastewater for spray irrigation may be particularly risky as this may facilitate virus attachment to produce surfaces (Richards, 2001). Green onions and other select produce items may be particularly prone to virus contamination because their surfaces are complex, allowing fecal matter and other organic materials to adhere tenaciously (Centers for Disease Control, 2003c). The survival of viruses on vegetables has been shown to be dependent on pH, moisture content, and temperature (Harris et al., 2002). Because noroviruses and HAV have been associated with a number of produce-associated outbreaks, it seems possible, though not yet supported by studies, that these viruses may be resistant to some of the virucidal substances naturally found in produce such as organic acids, and phenolic and sulfur compounds (Seymour and Appleton, 2001).

As items implicated in outbreaks are usually picked and processed long before consumption, it is often difficult to identify the point at which contamination occurred (Hutin et al., 1999). Moreover, locating the growing site of a particular produce item may be complicated. For example, in the case of an HAV outbreak linked to imported lettuce, the names of the farms supplying the lettuce were not included on the product labels, making it impossible to trace the geographic origin of the produce item (Rosenblum et al., 1990). When combined with issues such as poor patient recall and the extended incubation period for HAV, trace-back of contaminated product is very difficult (Calder et al., 2003; Fiore, 2004).

Items such as green onions, which have recently been implicated in HAV outbreaks in the United States, may become contaminated at any time during the production and processing continuum by contaminated soils, water, or human handling. However, as this particular produce requires extensive human handling during harvesting, it has been suggested that human handling is perhaps the most likely source of virus contamination (Dentinger et al., 2001). Several recent enteric virus outbreaks associated with the consumption of contaminated fresh produce are presented in Table 13.2.

3.1. Preharvest Control Strategies

The Guide to Minimize Microbial Food Safety Hazards for Fresh Fruits and Vegetables (U.S. Food and Drug Administration, 1998) provides a framework

Table 13.2 Epidemiological studies summarizing recent enteric virus outbreaks associated with produce items

<i>Agent</i>	<i>Food</i>	<i>Sample tested</i>	<i>Detection Methods</i>	<i>Conclusions</i>	<i>Reference</i>
HAV	Blueberries	Clinical and food	RT-PCR, Gel electrophoresis, Dot-blot hybridization, Sequencing	Contamination by infected food handlers or by faecally polluted groundwater	Calder et al., 2003
HAV	Green onions	Clinical	Serological testing	Contamination occurred in the distribution system or during growing, harvest, packing or cooling	CDC, 2003
HAV	Green onions	Clinical	RT-PCR, Sequencing	Contamination probably during harvesting	Dentinger et al., 2001
Norovirus	Lettuce	Clinical	Radioimmunoassay, EM	Unsanitary handling of lettuce or cross-contamination by raw seafood	Griffin et al., 1982
HAV	Strawberries	Clinical	RT-PCR, Sequencing	Contamination occurred during harvest due to unsanitary conditions	Hutin et al., 1999
HAV	Lettuce	Clinical	Serologic testing	Contamination occurred before local distribution	Lisa et al., 1990
HAV	Strawberries	Clinical and food	Immunoselection, RT-PCR, Hybridization	Contamination occurred probably by an infected picker	Niu et al., 1992
Norovirus	Raspberries	Clinical and food	RT-PCR, Hybridization, Sequencing	Contaminated water (irrigation or before packaging)	Ponka et al., 1999
HAV	Raspberries	Clinical	Immunoassay	Contamination at the picking stage	Reid, 1987

for the identification and implementation of practices likely to decrease the risk of pathogen contamination in fresh produce from production, packaging, and transport based on Good Agricultural Practices (GAPs) and Good Manufacturing Practices (GMPs). This document provides guidance for the proper management, handling, and application of animal manure. Emphasis should also be placed on assuring that waters used in production (for irrigation and pesticide application) are of high quality and do not present a human health hazard. There is, however, little conclusive data regarding the efficacy of sewage treatment on virus inactivation or on the degree of virus persistence in treated sewage or sludge. The virucidal efficacy of sewage disinfection can often be limited due to virus aggregation and association with particulate matter, and the occurrence of enteric viruses in sewage is usually sporadic. Estimates of the efficacy of secondary sewage treatment and disinfection on enteric viruses removal range from 1 to 2 orders of magnitude, while chlorination can remove an additional 1–3 orders of magnitude of enteric viruses depending on the dose, temperature, and contact time (Schaub and Oshiro, 2000). Unfortunately, sewage spills, storm-related contamination of surface waters, illicit discharge of waste, and residential septic system failures are widely recognized as the leading sources of surface water and groundwater contamination, which may impact fruit and vegetable production (Suslow et al., 2003). Scientific reports that document the feasibility and performance of various methods of on-farm water treatment (such as chlorination, peroxyacetic acid, UV, and ozone treatment) are also scarce. The risk associated with the reuse of wastewater for irrigation also requires further investigation (Gantzer et al., 2001).

Because many produce items are subjected to extensive human handling during harvesting, preharvest food safety strategies should also focus on food handlers. On-site toilet and hand-washing facilities should be readily accessible, well supplied, and kept clean. All employees (full-time, part-time, and seasonal personnel), including supervisors, should have a good working knowledge of basic sanitation and hygiene principles, including proper hand-washing techniques (FDA, 1998). The employees should be instructed to report any active cases of illness to their supervisors before beginning work (Koopmans et al., 2002). Furthermore, the presence of children at picking sites should be discouraged, and appropriate childcare programs should be available so that workers are not forced to bring their young children into the fields (Fiore, 2004).

3.2. Postharvest Control Strategies

Many produce items are washed before entering the distribution phase of the farm-to-fork chain. Washing fresh produce can reduce overall microbial food safety hazards so long as the water used in such rinses is of adequate quality. Of course, water (and ice) used for rinsing and packaging must originate from a pristine source or be decontaminated with chlorine or by some other disinfection method. However, washing and disinfection may not be sufficient to eliminate viral contamination from vegetables. Surface morphology and physiologic characteristics of the produce item(s) certainly com-

plicate disinfection efficacy; leafy vegetables can be more difficult to decontaminate because of their rough or wrinkled surfaces, and small fruits like raspberries and blackberries have more porous and complex surfaces that can entrap virus particles (Richards, 2001). When fresh produce is cut or damaged, viruses can be sequestered in abrasions. The most commonly used sanitizers for washing fruits and vegetables are chlorine, chlorine dioxide, and organic acids (Seymour and Appleton, 2001). Ozone has been put forth as a potential disinfectant but may be less promising because oxidation of food components may result in discoloration as well as deterioration of flavor. Toxicity and reactivity are other disadvantages associated with ozone.

3.2.1. Water, Produce Washes, and Household Chemicals

Produce items are frequently washed at numerous steps along the post-harvest continuum. A general rule of thumb is that water washing alone can remove about $1 \log_{10}$ of microbiological contaminants from the surface of produce items, keeping in mind that this estimate varies with factors such as produce type, virus type, degree of viral contamination, and water temperature. For instance, Lukasik et al. (2003) evaluated a variety of simple methods to remove viruses from a model produce commodity. Using strawberries artificially contaminated with poliovirus and bacteriophages MS2, Φ X174, and PRD1, these investigators found that water immersion and hand rubbing of the berries in water held at 22°C or 43°C resulted in removal or inactivation of 41–79% and 60–90% of the input virus, respectively. Overall, hand rubbing in water held at a higher temperature (43°C) facilitated virus removal. These same investigators also evaluated a commercial produce wash called Fit® (Proctor and Gamble) for its ability to reduce virus load in artificially contaminated strawberries and found that virus inactivation ranged between 80% and 90%. Finally, these authors reported that automatic dishwashing detergent (ADWD, 0.05%) and 10% vinegar were more effective than Healthy Harvest®, 0.05% liquid dishwashing detergent, or 2% NaCl for removal of viruses from strawberries immersed in lukewarm water. In this case, supplementing washwater with vinegar and ADWD produced virus reductions ranging from 95% to >99% (Lukasik et al., 2003).

3.2.2. Chlorine, Chlorine Dioxide, and Other Comparative Studies

There is a long history of using chlorine to control microbial contamination in water. Unfortunately, there is a paucity of published data regarding the efficacy of chlorine in the inactivation of viruses from the surface of produce items. In one recent study, Lukasik et al. (2003) reported on the efficacy of chlorine in inactivating viruses from inoculated strawberries. The levels of bacteriophages MS2, Φ X174, and PRD1 and poliovirus type 1 were reduced by 70.4% to 99.5% when strawberries were immersed for 2 min in 43°C water containing 0.3 ppm to 300 ppm of chlorine. Free chlorine at 200 ppm was considered optimal because it gave the same degree of inactivation as did 300 ppm free chlorine.

The HAV was reduced more readily (around 96%) when strawberries were washed in tap water supplemented with 2 ppm chlorine dioxide (ClO₂)

for 30 min, as compared with the same ClO_2 concentration used in wash water for a 30-s exposure period (around 67% inactivation). These results suggest that, under realistic processing conditions, chlorine dioxide washes are not very effective in reducing viral risks associated with this product (Mariam and Cliver, 2000).

In a study comparing the antiviral activity of commonly used antimicrobials (5.25% sodium hypochlorite, quaternary ammonium compounds, and 15% peroxyacetic acid–11% hydrogen peroxide) for rinsing produce, it was found that FCV could survive in strawberries and lettuce after they had been washed for 10 min at room temperature (Gulati et al., 2001). Only peroxyacetic acid–hydrogen peroxide formulations were proved to effectively reduce FCV titers by $3 \log_{10}$, although this occurred only at concentrations four times higher than those permitted by the FDA. In general, organic acids are unlikely to cause significant inactivation of enteric viruses, because viruses have mechanisms that facilitate their survival in the low acidity of the stomach. For example, HAV has been demonstrated to be extremely acid stable, remaining infectious after 5 hr of exposure to pH 1 at room temperature (Scholz et al., 1989).

In a recent study, trisodium phosphate (TSP; 1%), a common household cleaner, was found to be as effective as 0.5% hydrogen peroxide for the reduction of representative bacteriophages and poliovirus type 1 from artificially inoculated strawberries immersed in water held at 43°C; the inactivation rates ranged from 97% to >99% (Lukasik et al., 2003). A 0.5% solution of hydrogen peroxide, however, caused bleaching of the product and although this was ameliorated with a 10-fold decrease in concentration, the efficacy of the 0.05% peroxide solution was essentially the same as that of tap water washes alone. Cetylpyridinium chloride (CPC) was less effective at virus inactivation, ranging from 85% to 97% when applied to the surface of the strawberries (Lukasik et al., 2003).

Taken together, the data on chemical disinfection for the inactivation of viruses from food surfaces is not all that promising and is quite variable. For instance, disinfectants incorporated in the wash water may not be effective in removing or inactivating viruses that have penetrated through the skin of the produce or those that might have entered tissues through cuts and abrasions. An additional hurdle is that the surfaces and textures of fruits and vegetables may be rough, wrinkled (leafy vegetables), or porous (strawberries, raspberries, and blackberries), allowing the entrapment of viruses, thereby sequestering them from disinfectants. In general, washing produce items individually rather than in bulk is recommended, as bulk washing may result in the infiltration of viruses into produce items that were not initially contaminated, (Richards, 2001).

3.2.3. Alternative Decontamination Methods

Ultraviolet radiation has been suggested as an alternative to chlorine for water disinfection. FCV and poliovirus type 1 have proved to be highly susceptible to inactivation by UV radiation, with $3 \log_{10}$ reductions achieved by

doses of 23 and 40 mJ/cm², respectively (Gerba et al., 2002; Thurston-Enriquez et al., 2003). Bench-scale ozone disinfection of water using a dose of 0.37 mg of ozone/liter at pH 7 and 5°C resulted in at least a 3 log₁₀ reduction of norovirus and poliovirus type 1 within a contact time of 10 s. This promising technology may some day prove to be an alternative to chlorine for the disinfection of produce wash water. Although many novel disinfectant washes with effective antiviral properties are available at the consumer and processor levels, their use directly on produce surfaces is frequently prohibitive due to unacceptable organoleptic changes in the produce. Likewise, gamma radiation between 2.7 and 3.0 kGy has been shown to reduce HAV on lettuce and strawberry surfaces by 1 log₁₀. However, this dose currently exceeds the U.S. standards for the use of irradiation to control sprouting and pest infestation in produce items (Bidawid et al., 2000).

3.2.4. Temperature

Viruses can survive in contaminated fruits and vegetables under household refrigeration conditions (Kurdziel et al., 2001). The survival of poliovirus on the surface of foods has been demonstrated in many previous studies (Ansari et al., 1988; Mbithi et al., 1992; Abad et al., 1994). For example, poliovirus titers dropped by 1 log₁₀ after 12 days of refrigerated storage in lettuce and white cabbage, whereas on green onions and fresh raspberries its concentration remained unchanged under the same storage conditions. Similarly, a study on the persistence of HAV on fresh produce (lettuce, fennel, and carrot) demonstrated produce-specific variation in the ability of the virus to withstand refrigeration. More specifically, HAV survived on lettuce until the ninth day of refrigeration but decreased to undetectable levels after 7 and 4 days of refrigeration on fennel and carrots, respectively (Crocì et al., 2002).

In a multistate HAV outbreak associated with the consumption of frozen strawberries, it was apparent that the virus had survived storage at frozen temperatures for up to 2 years (Niu et al., 1992). In a laboratory-based study of frozen strawberries contaminated with poliovirus, the investigators found only 1 log₁₀ reduction in virus titer within the first 9 days of freezing (Kurdziel et al., 2001). Rotavirus SA-11 survived for almost a month on lettuce, radishes, and carrots when stored at refrigeration temperatures, while survival was significantly less when the produce was stored at room temperature (Badawy et al., 1985). In another study, 93% of the initial rotavirus contamination could still be recovered from lettuce after the inoculum was allowed to dry for 4 hr at room temperature (O'Mahony et al., 2000).

4.0. READY-TO-EAT FOODS

Ready-to-eat (RTE) foods are defined as those products that are edible without washing, cooking, or additional preparation by the consumer or by the food service establishment (Public Health Service, 1999). In general, this means that such foods are not subjected to a terminal heating step prior to

consumption. In RTE foods, transmission of enteric viruses through food handlers is widespread. Indeed, recent epidemiological surveillance data (1988 to 1992) indicate that poor personal hygiene of infected food-handlers was the most commonly cited factor contributing to food-borne outbreaks of HAV (96%) and norovirus-associated gastroenteritis (78%) (Bean et al., 1997). The cost of viral disease outbreaks due to infected food-workers can in some instances be very high because they frequently involve secondary transmission, and, especially for HAV, the cost of widespread prophylaxis is very high (Daniels et al., 2000).

4.1. The Epidemiological Significance of RTE Foods

4.1.1. The Role of Fecal-Oral Transmission

Food handlers may transmit viruses to foods from a contaminated surface, from another food, or from contaminated hands. The ultimate source of viral contamination is usually human fecal matter, although vomitus may also contain infectious virus. Because contamination of RTE foods occurs post-processing, no level of upstream food processing will control the problem (Richards, 2001). One of the major hazards for cooked RTE foods arises through handling with bare hands (Bryan, 1995). Technically, any RTE food handled by a symptomatic or asymptomatic virus carrier can become contaminated. However, certain food products have received considerably more attention (e.g., salads, raw fruits and vegetables, bakery products) over the years, probably due to their association with high-profile outbreaks. Recent enteric virus outbreaks associated with RTE foods are shown in Table 13.3.

Nonenveloped viruses, such as rotavirus, noroviruses and HAV, survive better on skin than do enveloped ones such as herpes and influenza viruses (Springthorpe and Sattar, 1998). There is strong evidence suggesting that contaminated hands frequently play a role in virus spread, acting as either virus donors or recipients. Hands can become contaminated by direct contact with any virus-containing fluid from self or others; they may also become individually contaminated by contacting virus-contaminated surfaces or objects (Sattar et al., 2002). The extent of such contamination will vary depending on a variety of factors, including the virus load, the degree of discharge from the host, the hand-washing habits of the infected person, and the efficiency with which virus is transferred and persists. Considerable amounts of HAV (16–30% of the initially recoverable virus) remained infectious on finger pads after 4 hr, even though 68% of viral infectivity was lost within the first 1 hr (Mbithi et al., 1992). In another study, rotavirus remained infectious on human hands for 60 min after its inoculation, and it could be transferred from the contaminated hands to animate and nonporous inanimate surfaces (Ansari et al., 1988). In fact, twice as much virus was transferred by the hand-to-hand route when compared with that transferred between hands and nonporous inanimate surfaces. Transfer studies with PDR-1 phage, used as a surrogate for human viruses, revealed that infectious particles could be transmitted under ordinary circumstances from the surface of fomites, such as

Table 13.3 Epidemiological studies summarizing recent enteric virus outbreaks associated with RTE foods

<i>Agent</i>	<i>Food</i>	<i>Samples tested</i>	<i>Detection Methods</i>	<i>Conclusions</i>	<i>Reference</i>
Norovirus	Salads	Outbreak samples (clinical, serum and food)	RT-PCR	Ill food handler contaminated the salads	Anderson et al., 2001
Norovirus	Box lunch	Outbreak samples (clinical)	RT-PCR EM	Person-to-person transmission	Becker et al., 2000
Norovirus	Bakery products	Outbreak samples (clinical and serum)	Immune-EM, Radioimmunoassay	Ill handler during the outbreak	Kuritsky et al., 1984
Norovirus	Turkey salad sandwiches	Outbreak samples (clinical and food)	EM, Immune-EM, bacteriological examination	Mechanical transmission of the virus or pre-symptomatic food handler	Lo et al., 1994
Norovirus	Deli sandwiches	Outbreak samples (clinical and food)	RT-PCR (single and nested) Sequencing Southern hybridization	Food handler slicing the ham had an ill infant	Parashar et al., 1998
Norovirus	Potato salad	Outbreak samples (clinical)	EM, RT-PCR	Kitchen assistant vomited in the sink, where the salad was later prepared	Patterson et al., 1997
HAV	Bread	Outbreak samples (clinical)	Serum and saliva tests for IgM and IgG	Handler with soiled hands contaminated samples when wrapping them	Warburton et al., 1991

telephone receivers, to the hands of a person using the receiver. If there were subsequent fingertip-to-mouth contact, infection might result (Rusin et al., 2002).

Hepatitis A, with an incubation period of 15–50 days, appears more readily transmitted during the latter half of the incubation period, meaning that food workers in retail settings with acute HAV infection can readily contaminate RTE products if they do not practice adequate personal hygiene (Daniels et al., 2000). Although adults are considered infectious only in the first few days of a norovirus infection, it has been shown recently that they can shed viruses in their feces for up to 2 weeks from disease onset (White et al., 1986; Yotsuyanagi et al., 1996; Parashar et al., 1998). Infected infants may be able to shed virus for more than 2 weeks (Daniels et al., 2000). Further complicating the issue is evidence of presymptomatic fecal excretion from food handlers while incubating the disease (Lo et al., 1994). Indeed, an outbreak has been documented in which a food handler was able to transmit calicivirus a few hours before becoming symptomatic (Gaulin et al., 1999).

4.1.2. The Role of Vomitus and Secondary Spread

Although the fecal-oral transmission route is the most important in promoting the spread of noroviruses, the role of vomitus cannot be overlooked. More than 30 million virus particles can be liberated from one vomiting episode, and when compared with an infectious dose of 10–100 particles, this is a significant virus load (Caul, 1994). The importance of this lies in its contribution to secondary spread, because aerosolization of vomitus can result in infection of exposed subjects who inhale and subsequently swallow the aerosolized virus (Marks et al., 2000). Air currents generated by air conditioning or open windows can disperse aerosols widely (Caul, 1994), while ceiling fans can also contribute to the virus spread (Marks et al., 2000). Evidence for respiratory spread has not been documented, and seems unlikely, as replication of noroviruses in respiratory mucosal cells does not occur (Lopman et al., 2002).

4.1.3. The Role of Fomites

Enteric viruses may persist for extended periods of time in foods and on materials and objects that are commonly found in institutions and domestic environments, including paper, cotton cloth, aluminum, china, latex, and polystyrene (Abad et al., 1994). Thus, viruses can be transmitted by mechanical transfer from the contaminated object (Lo et al., 1994). A recent outbreak of norovirus in an elder-care residential hostel in Australia is a case in point. In this case, the vomitus of an infected individual served as the source of virus that contaminated furniture and carpets. The virus remained infectious even after these items were professionally cleaned, serving as an intermediate source of virus transmission (Liu et al., 2003). In general, it is difficult to investigate whether and to what extent fomites assist in the spread of enteric viruses (Abad et al., 1994). Moreover, apart from more predictable surfaces like carpets and toilets seats, other surfaces such as lockers, curtains, and com-

modes have also been implicated in virus transmission in hospital outbreaks (Green et al., 1998).

4.2. Prevention Strategies

4.2.1. Decontamination of Hands

For hand decontamination to be successful in controlling viral food-borne disease outbreaks, three elements must be in place: (i) an effective disinfecting agent, (ii) adequate use instructions, and (iii) regular compliance (Sattar et al., 2002). Because hands are believed to play an important role in virus spread, the efficiency of several hand-washing agents has been investigated. In the first of such studies, Mbithi et al. (1993) showed that most surface disinfectants, even the alcohol-containing ones, were not able to eliminate poliovirus type 1 and HAV, based on an efficacy criteria of a $3\log_{10}$ reduction in virus titer (99.9% inactivation). A medicated liquid soap was the most effective against both viruses, although there were virus-to-virus differences in inactivation. Disturbing was the fact that as much as 20% of the initial virus inoculum could still be detected on hands after washing and drying, and nearly 2% of the input virus could be readily transferred to other surfaces. These investigators pointed to a need for establishment of new standards in the selection of effective formulations for hand-washing agents with respect to antiviral activities (Mbithi et al., 1993). Moreover, it is generally recognized that more work is needed to establish a standard hand-washing regimen upon which inactivation claims against viruses can be based for labeling purposes (Sattar et al., 2002). In a study of the efficacy of common hand disinfectants against a porcine enterovirus, all of the agents were proved ineffective with the exception of a 1% chlorine bleach solution (Cliver and Kostenbader, 1984). Ethanol-based hand rubs contributed to the reduction of FCV spread, but because they were not as effective as water and soap, the investigators suggested that they are perhaps more useful in the decontamination of hands between hand-washing events (Bidawid et al., 2004). A recent study suggested that contact for 30s with 1-propanol or ethanol solutions on hands could reduce FCV titer as much as $4\log_{10}$ (Gehrke et al., 2004). The same study indicated that an increase in disinfection effectiveness did not correlate with an increase in alcohol concentration, as alcohol-based solutions of 70% were more effective against FCV than were 90% solutions. This is in agreement with earlier findings reporting that a 70% alcohol formulation was effective for decontaminating rotavirus from hands (Ansari et al., 1989). The same group investigated the efficacy of aqueous solutions of chlorhexidine gluconate (Savlon and Cida-stat) in reducing rotavirus from hands, and the degree of virus removal was the same as that observed with tap water alone (Ansari et al., 1989).

Nearly 46%, 18%, and 13% of infectious FCV was transmitted from experimentally contaminated hands to ham, lettuce, and metal surfaces, respectively (Bidawid et al., 2004). On the contrary, less efficient virus transfer occurred from contaminated ham (6%), lettuce (14%), and metal sur-

faces (7%) to hands. In both cases, FCV transfer could be significantly interrupted if soiled hands were washed with water or both water and soap before contacting the recipient surface (Bidawid et al., 2004). Hand-washing with water was similarly effective in interrupting HAV transfer from contaminated hands to lettuce (Bidawid et al., 2000). The reduction of rotavirus from finger pads was approximately $3 \log_{10}$ better when using a gel containing 60% ethanol as a hand disinfectant than using a simple hard-water rinse (Sattar et al., 2000). Water rinsing after the application of the hand antiseptic agent followed by immediate drying can provide further reduction of viruses on washed hands (Ansari et al., 1989). Tap water is used in most studies for rinsing hands, although its composition may vary geographically and temporally. Moreover, organic and inorganic compounds in water may facilitate the removal of viruses from hands (Ansari et al., 1989). Residual moisture on hands after hand-washing has been found to play an important role in the transfer of viruses (Springthorpe and Sattar, 1998), meaning that air drying may be a critical step for virus removal, especially if the hand-washing agents are not very effective (Ansari et al., 1991). Hot-air drying of hands contaminated with porcine enterovirus type 3 was found to reduce the virus titer by 92% (Cliver and Kostenbader, 1984). A study by Ansari et al. (1991) found that, regardless of the hand-washing agent used, electric air drying produced the highest reduction in rotavirus when compared with either paper towels or cloth towels. For instance, after washing with soap and water and with no drying step, there was a 77% reduction of rotavirus on hands. On the other hand, a reduction of 92% was observed after warm-air drying compared with 87% and 80% virus removal using paper towel or cloth drying, respectively.

4.2.2. Decontamination of Surfaces

Contaminated surfaces can readily transmit viruses to hands or food upon contact. The survival of human enteric viruses on environmental surfaces depends on several factors, including temperature, relative humidity, type of surface, and virus type. The results of several surface inactivation studies are summarized in Table 13.4. HAV, for example, can remain infectious on nonporous inanimate surfaces for several days, and this survival is influenced by relative humidity and air temperature (Mbithi et al., 1991). In a large study, Mbithi et al. (1991) reported that the half-life of HAV ranged from more than 7 days at relatively low humidity and 5°C to about 2 hr at high humidity and 35°C. On the contrary, under the same experimental conditions, poliovirus type 1 survival was proportional to the level of relative humidity and temperature, with longer survival occurring at high relative humidity.

The persistence of human enteric viruses on environmental surfaces in the presence of fecal material has been investigated with results varying by both virus and surface. Abad et al. (1994) found that the survival of human rotavirus and HAV on surfaces was not affected by the presence of fecal material, while enteric adenovirus and poliovirus persistence on nonporous surfaces (aluminum, china, glazed tile, latex, and polystyrene) was enhanced

Table 13.4 Surface inactivation of viruses by common disinfectants

<i>Surface</i>	<i>Virus</i>	<i>Agent/Concentration</i>	<i>Contact time</i>	<i>Efficiency</i>	<i>Comments</i>	<i>References</i>
Polystyrene	HAV, HRV	Sodium chlorite,	1 min, 28°C	~3 log reduction	Presence of organic matter not increase virus persistence after disinfection	Abad et al., 1997
		Ethanol, Chlorhexidine digluconate, Sodium hypochlorite, Phenol and Sodium phenate, Diethylentriamine,				
Food-contact surface	FCV	5.25% sodium hypochlorite,	1 and 10 min, 22°C	>3 log reduction	Sodium hypochlorite: efficient only at 5,000 ppm available chlorine	Gulati et al., 2001
		1.75% iodine and 6.5% phosphoric acid, 3 quaternary ammonium compounds, 15% peroxyacetic acid and 11% hydrogen peroxide, 2 phenolic compounds				
Polyvinyl chloride, High-density polyethylene, Aluminum, Stainless steel, Copper	HAV	10% quaternary ammonium and 5% glutaraldehyde,	1 or 5 min, 4 and 22°C	2-7 log reduction	Efficiency increases at 22°C, 5 min contact time and concentration of 3,000 ppm of the active ingredient	Jean et al., 2003
		12% sodium hypochlorite, 2.9% dodecylbenzene sulfonic acid and 16% phosphoric acid 10% quaternary ammonium, 2% iodine, 2% stabilized chlorine dioxide				
				<3 log reduction		

Table 13.4 *Continued*

<i>Surface</i>	<i>Virus</i>	<i>Agent/Concentration</i>	<i>Contact time</i>	<i>Efficiency</i>	<i>Comments</i>	<i>References</i>
Stainless steel disks	HAV	2% gluteraldehyde, quaternary ammonium compound (with 23% HCl and sodium hypochlorite with >5,000 ppm chlorine), phenolics, iodine-based products, alcohols, solutions of acetic, peracetic, citric and phosphoric acids	1 min	>3 log reduction <3 log reduction	Only 2% gluteraldehyde, a Quat compound with 23% HCl and sodium hypochlorite with >5,000 ppm available chlorine are effective	Mbiti et al., 1990
Stainless steel disks	rotavirus	0.1% o-phenylphenol and 79% ethanol, 7.05% quat diluted 1:128 in tap water 6% sodium hypochlorite diluted to give 800 ppm free chlorine, 14.7% phenol diluted 1:128 in tap water	10 min	>4 log reduction <1 log reduction <2 log reduction	Only the disinfectant spray reduced virus infectivity more significantly than tap water alone (<1 log reduction)	Sattar et al., 1994

by the presence of feces. The persistence of the latter two viruses was unaffected, however, by the presence of fecal matter when deposited on porous (paper and cotton cloth) surfaces. Other important findings from this study focused on the ability of each virus to survive when dried on a surface. For example, the reduction of HAV and human rotavirus infectivity when placed on several fomites and dried for a period of 3–5 hr was not as significant as it was for adenovirus and poliovirus, implying that the former two viruses may be more likely to be transmitted after substantial environmental persistence. In another study, rotavirus SA-11 suspended in a stool preparation could be detected on contaminated environmental surfaces after 60 min of drying, while the same virus survived for only 30 min when suspended in water (Keswick et al., 1983). Similarly, cell culture–adapted human rotavirus was found to be significantly protected from drying when it was in a 10% fecal suspension rather than in distilled water (Ward et al., 1991). The presence of fecal material not only increases virus survival but has also been shown to protect poliovirus from the action of disinfectants (Hejkal et al., 1979). In fact, a fourfold increase in residual chlorine was required to achieve the same degree of inactivation for poliovirus type 1 suspended in feces as compared with a suspension of free virus at pH 8 and 22°C.

Cleaning and disinfection of surfaces are of major importance in the prevention of enteric viral disease. In general, there is a paucity of information on the efficacy of most commercial disinfectants against enteric viruses. Comparing a range of disinfectants used on experimentally contaminated polystyrene surfaces under conditions suggested by the manufacturer, only sodium chlorite proved to be effective against HAV and human rotavirus (Abad et al., 1994, 1997a). Work done with several commercial disinfectants used in the food industry showed that only products containing glutaraldehyde and sodium hypochlorite were effective in HAV inactivation, and their efficacy improved when the compounds were used at high concentrations and for a relatively long time (Jean et al., 2003). A majority of chemical disinfectants used in both institutional and domestic environments do not effectively inactivate HAV (Mbithi et al., 1990). Of the 20 formulations tested, only 2% glutaraldehyde, a quaternary ammonium compound containing 23% HCl, and sodium hypochlorite with free chlorine in excess of 5,000 ppm had demonstrable virucidal efficacy. These results supported the use of sodium hypochlorite for surface disinfection and were validated in a more recent study using FCV as a norovirus surrogate (Gulati et al., 2001). From a variety of disinfectants used at manufacturer-recommended concentrations, only sodium hypochlorite at 5,000 ppm available chlorine (200 ppm of chlorine is the FDA allowable level) was effective in reducing more than $3\log_{10}$ of FCV. In another study, the efficacy of commercially available disinfectants was tested and the most suitable for environmental surfaces was reported to be freshly prepared hypochlorite solution at high concentrations (1,000 ppm) (Doutree et al., 1999). In a transfer study, Sattar et al. (1994) reported a $4\log_{10}$ inactivation of rotavirus transfer from stainless steel disks to fingerpads of volunteers using a disinfectant spray (0.1% *o*-phenyl phenol and 79% ethanol), while domestic bleach (6% sodium hypochlorite diluted

to give 800 ppm free chlorine) and a phenol-based product (14.7% phenol diluted 1:128 in tap water) provided reductions of almost $3 \log_{10}$ of virus infectivity. No detectable virus was transferred to fingerpads from disks treated with these three agents, but when the disks were cleaned with tap water or a quaternary ammonium-based product, the transfer rates were 5.6% and 7.6%, respectively. In another study, Sattar (2004) tested the anti-FCV activity of various microbicides and discovered that the most effective one at the shortest contact time (1 min) was domestic bleach (5% sodium hypochlorite, 1,000 ppm available chlorine), which reduced the titer of FCV by nearly $4.5 \log_{10}$.

4.2.3. Education, Training, and Supervision

Fingers, and particularly fingernails, are thought to be the most important part of the hand in terms of the transfer and spread of pathogenic microflora (Lin et al., 2003). Fingernails are of particular concern because fecal material may be readily deposited in this location and is subsequently difficult to remove. This may be particularly important for those having long or “artificial” fingernails. A recent study demonstrated that the most effective way to remove virus from artificially inoculated fingernails was to scrub them with a nailbrush using soap (regular or antibacterial) and water. Alternatively, employees should be encouraged to maintain short nails, as these are less likely to harbor fecal material than long ones (Lin et al., 2003).

Indeed, food employees should not touch exposed RTE food with their bare hands and should use suitable items such as deli tissue, spatulas, tongs, and single-use gloves (smooth, durable, and nonabsorbent) or dispensing equipment (FDA, 2001). Gloves are the only FDA-approved barrier method allowable to date. Issues impacting the efficacy of gloves in preventing viral disease include the glove material, glove permeability, duration of wearing, and hand-washing techniques prior to and after wearing. Glove leaks are more frequent with vinyl than with latex gloves (Guzewich and Ross, 1999), and frequent replacement of disposable gloves is encouraged, particularly when gloves get damaged or soiled, or when interruptions occur in the operation. Where food contact by handlers is unavoidable, careful practices such as frequent hand-washing and prevention of cross-contamination during handling and preparation are suggested. Apart from the “no bare-hand contact with RTE” policy in the Food Code, there is no direct information on the effectiveness of hand hygiene and gloving regimens in the food industry (Paulson, 2003).

It is of major importance that food-handlers, including seasonal workers, should have appropriate health and hygiene education. Such training should also cover the potential risk of enteric virus transmission due to sick children in the household (Koopmans and Duizer, 2004). If a food preparation staff member reports a diarrhea or vomiting episode while at work, he or she should not be allowed to enter the kitchen again. All of the food handled by that worker, as well as any other food that may have been exposed to aerosolized vomitus, should be destroyed (Lo et al., 1994). Potentially con-

taminated surfaces in the kitchen should be thoroughly decontaminated with a freshly prepared hypochlorite solution that releases 1,000 ppm of available chlorine. Frequently handled objects such as taps and door knob should also be decontaminated, and bleach-sensitive items should be cleaned with detergent and hot water (Chadwick et al., 2000). Managers of food manufacturing, catering, and food service industries should restrict ill food-handlers from working directly with food or food equipment and provide a sick leave policy that allows workers to stay home while ill (Centers for Disease Control, 2003b). Moreover, as soon as food handlers return to work, they should be instructed that they still may be shedding virus for a period of days to weeks after symptoms have abated and that they should continue practicing stringent personal hygiene (Koopmans and Duizer, 2004).

Increased awareness of the risk of gastrointestinal disease due to virus transmission is encouraged among food safety professionals and the public. However, rapid control of viral gastroenteritis outbreaks can be difficult. For instance, while transmission via contaminated food or water may sometimes be prevented or contained, the potential for person-to-person spread cannot be eliminated. This is especially challenging for settings where close contact inevitably occurs (i.e., university dormitories) and especially where there are a number of susceptible people in confined quarters (hospitals, nursing homes, daycare centers) (Kilgore et al., 1996). The failure to recognize and report HAV among children in daycare facilities is an important and contributing factor in disease propagation (Gingrich et al., 1983). Viruses can be readily transmitted in daycare settings; however, it is the “silent” transmission through poor personal hygiene of food handlers that is becoming increasingly recognized (Sattar et al., 2002).

For gastroenteritis outbreaks in hospitals and nursing homes, efforts to control virus circulation in the environment by immediate isolation of the case(s) should be undertaken. The timing of the last cleaning process should ideally be at least 72 hr after resolution of the last case (Chadwick et al., 2000). Hypochlorite is generally not recommended for the disinfection of carpets; however, steam has been suggested as a viable alternative (Cheesbrough et al., 1997; Chadwick et al., 2000). Indeed, during a viral gastroenteritis outbreak in a hotel, the carpets appeared visually clean after cleaning with detergent and vacuuming, but nonetheless they remained contaminated with infectious norovirus (Cheesbrough et al., 2000).

4.2.4. Vaccination

Immunity to HAV confers complete protection against reinfection. To date, there are three FDA-licensed HAV vaccines on the market. These are generally administered as a single primary immunization, followed by a booster dose 6–12 months later (Lemon, 1997). The vaccine efficacy is 94–100% and protection is likely to last for more than 20 years after vaccination (Fiore, 2004). Hepatitis A vaccination has been limited to high-risk groups and is currently approved for use in the United States in children over 2 years of age. Routine vaccination of all food handlers as a pre-exposure prophylaxis

is not recommended because their occupation does not put them at unusually high risk of infection. Furthermore, vaccinating all restaurant employees is unlikely to happen, as the cost to restaurant owners often exceeds the perceived benefits, even during a hepatitis A epidemic (Meltzer et al., 2001). However, some believe that HAV vaccination should be routinely available to people with increased occupational risk such as food handlers, health care workers in infectious disease and pediatrics sections, medical staff in laboratories handling stool samples, staff in daycare centers, and sewage treatment plant workers (Hofmann et al., 1992).

Immunoglobulin (IgG) is not the recommended choice for pre-exposure prophylaxis as it provides only short term (1–2 months) protection from HAV infection (Fiore, 2004). However, postexposure prophylaxis with IgG has been shown to be effective in eliminating or reducing the severity of hepatitis A infection, provided it is administered within 2 weeks of exposure and not within the late incubation period of the disease (Pavia et al., 1990). In the case of food-related exposures, it is recommended that IgG postexposure prophylaxis should be given to all food handlers, even if only one handler in that facility has been diagnosed with HAV infection. Moreover, IgG is recommended for the patrons of that establishment provided all of the following considerations exist: (i) the infected handler was responsible for handling RTE foods and was not wearing gloves, (ii) the infected handler had poor hygiene practices or had diarrheal symptoms, and (iii) the patrons can be identified and treated within 2 weeks of exposure (Committee on Infectious Diseases—American Academy of Pediatrics, 1991). The efficacy of administering HAV vaccine for postexposure prophylaxis remains to be established. It has been demonstrated that anti-HAV seroconversion occurs 14 days after vaccination and the average incubation period for HAV infection is 28–30 days, suggesting that vaccination may be protective if given within a few days of exposure (Koff, 2003).

A safe and effective vaccine against noroviruses would reduce the burden of the disease, which may be of particular importance for controlling gastroenteritis in children of developing countries, partly because repeated diarrheal episodes can cause damage to the intestinal mucosa leading to the development of malnutrition (Kapikian et al., 1996). Recent human challenge studies with the noroviruses have demonstrated both short-term and long-term immunity (Johnson et al., 1990; Parrino et al., 1977). The distinct epidemiological patterns of the noroviruses have introduced some technical difficulties for vaccine development. For instance, there may be multiple antigenically and genetically distinct strains of noroviruses circulating at any one time, and there is evidence indicating that infection with one strain does not provide cross-protection against other strains. This virus genus remains noncultivable, so it is difficult to routinely determine the presence of neutralizing antibodies in an infected individual (Estes et al., 2000; Hale et al., 2000). Recently, specific histological blood group antigens have been identified as putative ligands for the attachment of different norovirus strains to mucosal cell surfaces (Harrington et al.,

2004; Hutson et al., 2004). These observations require further investigation to determine whether there is any correlation between individual differences in susceptibility and specific virus genotype and/or the genetic background of the host (Harrington et al., 2004). The production of recombinant norovirus capsid protein and its formulation as an oral vaccine is an alternative approach to vaccination; however, the efficacy of this type of approach has yet to be established (Estes et al., 2000).

5.0. CONCLUSIONS

For all food commodities, preventing direct contact with human fecal material (and in some instances, direct exposure to vomitus) is obviously the first consideration in controlling virus contamination. Prevention of sewage disposal in harvesting waters is critical in controlling viral contamination of shellfish. However, the lack of correlation between the fecal coliform index and the presence of enteric viruses may at times complicate the ability of regulators to recognize contamination when it occurs. For produce items, adherence to GAPs, including attention to personal hygiene of field workers, is essential to controlling contamination at the preharvest phase. Likewise, proper personal hygiene, including the use of barrier protection and appropriate hand and surface decontamination, provides a first line of defense for preventing viral contamination of RTE foods. A critical consideration is providing food handlers with appropriate and ongoing education in hygienic practices. Designing effective educational programs for food handlers is notoriously difficult, as this itinerant population may have limited English language fluency, generally has lower educational attainment, and turns over quite rapidly.

It is clear from this discussion that low temperatures (refrigeration and freezing) are not reliable means by which to reduce enteric viruses in contaminated foods. High temperature (heating) may be effective in some instances, but recommended time-temperature combinations are virus-specific and will vary with the food commodity. A common theme for the food items discussed here is that most do not undergo a terminal heating step before consumption, so the relevance of heating may be limited simply by virtue of the specific commodity.

The efficacy of other postharvest controls is also somewhat limited, and, in general, these may reduce but will not eliminate viral contamination in foods. This is the case with depuration, relaying, and ionizing radiation as applied to raw, molluscan shellfish. High hydrostatic pressure appears to be a more promising technology, but much more data are needed to definitively establish its efficacy in inactivating viruses in this commodity. Likewise, washing produce with fresh, clean water, both with or without the addition of chemical disinfectants, may also reduce virus load in contaminated items, but it is important to note that the efficacy of such decontamination varies with both virus and produce type.

Widespread vaccination may eventually become an effective control measure for HAV, but it is not likely that norovirus vaccination will be a reality in the near future. It must also be noted that, once an outbreak occurs, strict infection control measures must be instituted to prevent further virus dissemination. Indeed, there are many opportunities for future research in prevention and control. Specifically, research is needed to identify effective intervention and control strategies, to develop improved monitoring and detection methods, to expand immunization options, and to develop successful food-handler educational programs. Working together, scientists can make further inroads in the prevention and control of viral food-borne diseases into the future.

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