Journal of Industrial Microbiology & Biotechnology (2001) 27, 117–125

www.nature.com/jim

FOOD-BORNE PATHOGENS

Enteric virus contamination of foods through industrial practices: a primer on intervention strategies

GP Richards

U.S. Department of Agriculture, Agricultural Research Service, Delaware State University, W.W. Baker Center, Dover, DE 19901, USA

Hepatitis A and E viruses, rotaviruses, Norwalk-like caliciviruses, and astroviruses are among the enteric viruses known to cause food- and waterborne illness. These viruses are spread by the fecal–oral route and are a major cause of morbidity and mortality worldwide. Foods may be contaminated at any time pre- or post-harvest; however, many outbreaks are associated with foods handled by infected restaurant workers. Produce may be contaminated by improper irrigation or fertilization practices, by the hands of infected pickers or processors, or as the result of adulteration during any stage of handling. Outbreaks have been commonly associated with foods which are served raw or only lightly cooked, such as molluscan shellfish, fruits and vegetables, and salads or products contaminated after cooking like frosted bakery products. The farming, shellfish, processing, transportation, and restaurant industries must maintain vigilance to reduce outbreaks of enteric virus illness. Intervention strategies to enhance product safety include increased industry and consumer education; changes in industrial practices, product management, and processing technologies; worker immunizations; and the development of improved monitoring tools for the detection of enteric viruses in foods. *Journal of Industrial Microbiology & Biotechnology* (2001) **27,** 117–125.

Keywords: Norwalk virus; hepatitis A virus; rotavirus; foodborne illness

Introduction

Enteric viruses comprise a broad assortment of viruses that are transmissible to humans through contaminated food and water and through person-to-person contamination. Among the most notable foodborne enteric viruses are hepatitis A and E viruses, small round-structured viruses, and group A rotaviruses. All of these viruses can be spread readily to foods during food production, processing, and handling. Hepatitis A virus has contributed to numerous foodborne outbreaks and is often associated with raw or lightly cooked shellfish [83]. The largest foodborne outbreak of hepatitis A was from the consumption of contaminated clams where nearly 300,000 people became sick during a 3-month period [38]. In the United States, hepatitis A causes an estimated 83,000 illnesses per year [70]. To date, there have been only two reported cases of domestically acquired hepatitis E in the United States [29,56]. Epidemics of hepatitis E have been reported in Asia, Africa, and Mexico [19]. Both of these viruses can migrate from the intestinal tract to infect and damage the liver, leading to jaundice and, occasionally, death. Hepatitis A is often self-limiting and spontaneous recovery is common [88]. The fatality rate from hepatitis A is about 100 per year in the United States [57]. Hepatitis E has a low mortality rate for the general population (0.5–3.0%); however, pregnant women have a mortality rate between 15% and 25% [64]. The incubation period for hepatitis A is 25–45 days, whereas the incubation period for hepatitis E is 15–60 days.

Caliciviruses, small round-structured viruses, may be divided into three genogroups represented by Norwalk virus, Snow Mountain virus, and Sapporo virus. Genogroup I viruses include Norwalk, Southampton, and Desert Shield viruses. Genogroup II includes the Snow Mountain, Camberwell, Lordsdale, Mexico, Toronto, Bristol, Hawaii, and OTH-25 viruses. Collectively, genogroups I and II viruses may be referred to as Norwalk-like viruses. Genogroup III consists of the classical caliciviruses with Sapporo virus as the prototype. Other members of this genogroup include Houston, London, and Parkville viruses. The Centers for Disease Control and Prevention recently estimated the annual incidence of Norwalk-like viral illness in the United States at 23 million [70]. The caliciviruses may cause diarrhea and vomiting with rapid onset, but lasting for only a few days. Norwalk-like viruses have been listed as the leading cause of foodborne hospitalizations in the United States today and responsible for 7% of the food-related deaths [70].

Rotavirus illness, also referred to as acute infantile diarrhea, is a common cause of infantile diarrhea worldwide. The exact role of foods in rotavirus transmission has not been elucidated; however, food, water, and person-to-person are all vehicles of transmission. It was estimated that only 1% of rotavirus cases was foodborne [70]. Rotavirus diarrhea may lead to dehydration and death, particularly in regions of the world where rehydration therapy is not available. Rotavirus causes an estimated 2.7–3.9 million illnesses, 49,000–50,000 hospitalizations [9,70], and about 30 deaths per year in the United States [70], but an estimated 800,000 deaths per year worldwide [76]. Although rare in the United States, rotavirus deaths are common in poor countries where rehydration therapy is unavailable.

Other enteric viruses include the astroviruses, enteric adenoviruses, and coronaviruses, which are also spread by the fecal-to-oral route. The number of astrovirus infections in the United States is estimated at 3.9 million per year with only 10 deaths [70]. Water

Correspondence: Dr GP Richards, USDA, ARS, Delaware State University, W.W. Baker Center, Dover, DE 19901, USA
Received 23 June 2000; accepted 22 September 2000

and person-to-person transmissions may be responsible for the majority of illnesses from hepatitis A, rotavirus, and astroviruses; but 40% (about 9.2 million cases) of Norwalk-like virus illnesses may be foodborne [70]. The number of enteric virus illnesses in the United States can only be estimated because most of the illnesses are mild, go unreported, and routine testing of patients for specific virus infections is not performed.

Unlike some bacterial pathogens which may require a moderate to high dose to induce illness, it is believed that just a few virus particles may be sufficient to elicit infection. Viral contaminants may persist in the environment, on food surfaces, or within foods for extended periods [25,94,96]. Hepatitis A virus was detected in cream cookies maintained at 21°C and 49°C for up to 30 and 14 days, respectively, in oysters maintained at 12–24°C for >5 days and in environmental samples (soil and water) at 25°C for 56–84 days [94]. Fortunately, human enteric viruses can only replicate in susceptible mammalian hosts, so there is no virus amplification within foods.

Enteroviruses consist of poliovirus, echovirus, coxsackievirus types A and B, and enterovirus serotypes 70 and 71. Poliovirus has been used extensively as a laboratory model since it is similar to hepatitis A virus in some respects, but unlike hepatitis A virus, it can be readily propagated in cell culture. The use of poliovirus has provided much information on the effectiveness of various treatments and processes for the disinfection of foods and food contact surfaces. Nearly all of the polioviruses present in the environment are vaccine strains which enter the environment from the stools of children recently immunized with an attenuated oral poliovirus vaccine. This vaccine is being phased out and the inactivated, injectable vaccine is being employed to remove all traces of poliovirus from the environment. Currently, polio has been eliminated from the Western Hemisphere and there is substantial progress being made to eradicate the disease worldwide. Over the next few years, it is likely that polio will be eradicated and that laboratory strains, even attenuated vaccine strains, will have to be destroyed. Soon, the poliovirus model will be a thing of the past. Unfortunately, there is no good successor for poliovirus; however, feline calcivirus is becoming a popular model for research on Norwalk–like viruses [93].

Pre-harvest contamination

Determination of the source of contamination is fundamental to development of an effective remedy. Pre-harvest contamination may occur in foods subjected to irrigation with reclaimed wastewater, crop fertilization with sewage sludge, or fecal pollution of the areas in which food products are obtained [14,75,90]. Non-point sources of pollution, like land runoff into watersheds and discharge of human wastes from boats, have caused major pollution events leading to foodborne illness [8,13,54,67]. Improperly operated sewage treatment plants, plants overloaded with storm water, or damaged or poorly maintained septic systems have been linked to outbreaks of enteric virus illness [73,99]. Land disposal of sewage sludge may contaminate groundwater and lead to potential disease outbreaks [96]. Improper dumping of septage or the leaching from baby diaper and related wastes from sanitary landfills can contribute to contamination of groundwater, which may in turn be used for drinking or irrigation. Dumps also contain insects, rodents, birds, raccoons, and other pests which may carry viruses many miles from the original disposal site. It is uncertain as to what extent wind and evaporation contribute to virus dispersal, although the incidence of illness among farm workers using spray irrigation of wastewater appears low [33,59]. During harvest, foods may be contaminated by harvesters, as was suspected in outbreaks of hepatitis A from strawberries [7,46,74], raspberries [82], and lettuce [89]; rotavirus from lettuce [41]; and Norwalk-like illness from raspberries [35] and oysters [54,67].

Post-harvest contamination

Enteric virus outbreaks have been associated with post-harvest contamination from the hands of food handlers, recontamination after cooking or processing, and inadequate sanitation. The restaurant industry has contributed to the incidence of illness due to poor managerial practice. Infected workers handling foods may transmit hepatitis A virus, Norwalk-like virus, and other enteric viruses to foods and no level of food processing will eliminate post-processing contamination. Virtually any food handled by ill workers may become contaminated, although certain food products have received more attention than others. Among the foods most commonly cited as contributing to viral illness from handling by infected workers are salads, raw fruits and vegetables [36,52,61,62,78], and bakery products [103,107] — foods that may be handled frequently and are not generally heated or reheated after handling. Cross-contamination of processed products by raw materials, like the contamination of salad by raw seafood, has also been reported [36]. Adequate plant sanitation may reduce the threat of product cross-contamination. Water must be of sanitary quality if it is to be used in food processing as an added ingredient in food preparation, or for use in manufacturing ice. Celery contaminated with non-potable water was responsible for an outbreak of Norwalk virus gastroenteritis, causing morbidity in over 1400 people [105]. Ice may become contaminated during or after production and can lead to food- or water-borne illness [12,51,58].

Industries at risk

Although most food-related industries are conscientious in producing safe and wholesome products, some are at higher risk of producing tainted products by the very nature of the food or the production process. Foods served raw or only lightly cooked are at greatest risk of transmitting viral contaminants obtained from either pre- or post-harvest sources. The molluscan shellfish industry is particularly susceptible since: (i) shellfish can bioconcentrate viruses within their edible tissues to levels much higher than in the water itself; (ii) there are no good methods to ascertain whether the shellfish or their harvest waters contain infectious viruses; and (iii) shellfish are frequently eaten raw or only lightly cooked. Outbreaks of foodborne viral illness have been attributable to virus-contaminated shellfish [20,83]. For the fruit and vegetable industries, farming practices, including fertilization and irrigation techniques, product transportation, and the health of the harvesters themselves, are coming under increased scrutiny. Today, food processors have an extensive list of disinfectants for sanitizing processing surfaces, but many disinfectants are ineffective in inactivating enteric viruses. Processors may also be unfamiliar with the best methods to apply disinfectants. There are many questions concerning processing conditions necessary to fully eliminate virus contaminants and further research is needed in this area. The

transportation industry must share the responsibility of maintaining clean trucks, ice, boxes, pallets, *etc.*, to haul food products. It is fortunate for food transporters that human enteric viruses do not replicate in foods even under abuse temperatures. The transportation industry should follow the same safeguards as for protecting foods from external sources of bacterial contamination. Perhaps, the segment of the economy most at risk from virus contamination of foods is the restaurant industry. They are impacted most because they have a greater potential for contaminating foods due to extensive handling, often by inexperienced and untrained workers. Restaurants may be held liable in civil and criminal prosecutions if their products cause illness. The restaurant trade is, by no means, the only industry culpable in the event of an outbreak, so all facets of the production from the growth, harvest, and distribution, to the processing and marketing industries must be accountable for their role in ensuring safe foods. Hotels and restaurants reportedly have a high level of virus transmission through person-to-person contacts [68]. Cruise ship lines have also been hit hard by enteric virus-like illness [42,43,51,69]. Outbreaks often involve a substantial number of passengers and crew. In these cases, transmission is not only from direct consumption of contaminated foods, but likely from person-to-person spread.

Intervention strategies

There are certain fundamental industrial practices that will allow the elimination or significant reduction of viruses from foods. Methods for elimination of enteric viruses from foods depend somewhat on the type of virus present and the food contaminated. No one step will be sufficient to ensure a safe and wholesome product since recontamination may occur at any stage in the food chain; however, a concerted effort will reduce the risk of foodborne illness. The following are some strategies that the food industry may consider adopting.

Industrial practices

The practice of using wastewater for crop irrigation and sewage sludge for fertilization should be evaluated carefully. One study demonstrated the ineffectiveness of anaerobic mesophilic digestion at 35–36°C to inactivate rotavirus and coxsackie B5 viruses; however, anaerobic thermophilic digestion at 54–56°C and aerobic thermophilic fermentation at 60–61°C inactivated these viruses [95]. Wastewater should be subjected to, at least, primary and secondary treatments followed by chlorination or ultraviolet (UV) light disinfection to reduce virus levels. Wastewater irrigation may pose a particular risk for produce that is consumed raw. Viruses from spray irrigation may be harbored on the surfaces of fruits and vegetables and can lead to illness if the produce is not adequately washed or peeled. An antibacterial compound for use in washing and disinfecting the surfaces of fruits and vegetables was recently marketed and could offer some benefits in reducing the levels of external viral contamination.

The harvest of shellfish should be strictly regulated in regard to the criteria of the National Shellfish Sanitation Program and the water quality criteria set forth by the Interstate Shellfish Sanitation Conference [100]. Under this program, shellfish-growing waters are classified based on the levels of coliform bacteria in the water. Although the fecal coliform standard does not correlate directly with the levels of viruses in the water or in the shellfish, outbreaks of enteric virus illness are not often reported for shellfish obtained from approved harvesting waters. The shellfish industry and regulatory agencies should continue to actively pursue shellfish poachers who obtain shellstock from closed areas. Sacks of shellfish should be clearly marked with tags providing information required by state health authorities regarding the date and location of the harvest and the name of the harvester. Such information is crucial in epidemiological studies designed to track and remove from the market foods implicated in outbreaks.

Sick food handlers have been responsible for numerous cases of foodborne viral illness [24,78,79,108]. All food industries should mandate that sick workers do not handle foods, packaging materials or ingredients, or processing line equipment, dishes or utensils during or immediately after their illness. Workers who have seemingly recovered from an illness may still carry and shed viruses which can lead to food contamination [108]. Employers should require that sick workers not be allowed to work until they are better, and the employees should be given temporary jobs away from the food for a few days after recovering from enteric illness. Provisions should be made to financially help sick employees, through paid sick leave or by other means, so that the workers will not hide the fact that they are sick for fear of a reduced paycheck.

Product and process management

Viruses may be present in seemingly clean foods. Since viruses are not associated with product spoilage, flavor and odor changes that signal spoilage from bacteria will not indicate the virological quality or safety of foods. Therefore, it is important to provide a system of checks and balances to ensure the safety of all foods. The food industry has subscribed to a new approach to protect the nation's food supply known as the Hazard Analysis Critical Control Point (HACCP) system [10,16]. Under the HACCP system, food operations must develop and implement a plan, based on sound scientific data and reasoning, to identify those points in the operation most likely to contribute to product contamination. These become the critical control points for the operation. The HACCP approach also requires that the critical control points be monitored to ensure proper operation of the system. In the case of cooked products, cooking times and temperatures would be monitored to ensure adequate inactivation of microbes. For canning operations, times and temperatures are routinely monitored to ensure that they are adequate to eliminate the presence of *Clostridium* spores. Similar safeguards are needed to control and eliminate viral contaminants. Many factors go into the development of a HACCP plan, but all too often, the health status of the employee is highly subjective. To reduce foodborne illness, employee health must be a prime consideration for all sectors of the food industry. Epidemiological data should also be used to design HACCP and risk assessment systems [81]. Over the next few years, HACCP is likely to expand into new areas of the food industry to encompass all aspects of the farm-to-fork continuum.

One fundamental attribute of the HACCP system is the monitoring aspect, where data and records are maintained and scrutinized for various critical control points. Under many HACCP programs, bacteriological tests are required to ensure that adequate processing times and temperatures are maintained or to demonstrate adequate sanitation of the equipment. Unfortunately, routine testing of viral contaminants in foods is not possible because virus assay procedures are generally lacking. In the absence of sound methods for virus detection, it may be necessary to identify potential hazards

not empirically identifiable. As improved detection methods become available for viruses, critical control points for viral contaminants may be identified.

Processing technology

Industrial application of processing technologies to reduce or eliminate viral contamination from foods can be highly effective, depending on the food type and the technology applied. Some of the processes that are effective in preventing, reducing, or eliminating enteric viruses in foods are discussed below.

Food disinfection practices

Disinfection and sanitation

Good sanitation can reduce the incidence of foodborne contamination. Sanitation guidelines should be in place for foods from production and harvest to the consumer. Current HACCP systems are beneficial in identifying and controlling those factors critical to the maintenance of safe products.

A number of studies determined the effectiveness of disinfectants in eliminating enteric viruses (Table 1). References from Table 1 should be scrutinized closely to determine if the disinfectants are appropriate for specific applications. Chemical disinfectants should be selected based on their effectiveness against foodborne viruses. Although solutions containing chlorine are effective disinfectants for most viruses, other disinfectants may be more practical or have residual effects to enhance sanitation efforts. In one study evaluating 27 chemical disinfectants for their ability to inactivate rotavirus on glass, plastic, and stainless steel surfaces, only nine were effective: chloramine-T, chlorhexidine gluconate, glutaraldehyde, hydrochloric acid, isopropyl alcohol, peracetic acid, povidone iodine, quaternary ammonium compound, and sodium-*o*-benzyl-*p*-chlorophenate [60]. The treatment of Norwalk virus with up to 10 mg/ml of chlorine for 30 min was insufficient to inactivate all the viruses present as determined by a volunteer study [50], although there is some speculation that the amount of free chlorine available was substantially less than 10 mg/ml. In another study, chlorine, chlorine dioxide, peracetic acid, and ozone were effective in reducing poliovirus, echovirus, coxsackievirus type B5, and rotavirus [39]. Rotavirus was readily inactivated by chlorine dioxide at alkaline pH values [22] and by chlorine [102] and ozone [101], particularly at acidic and neutral pH values. Chlorine dioxide and iodine, at alkaline pH values, have also been effective in inactivating poliovirus [3]. Engelbrecht *et al* [28] demonstrated that poliovirus types 1 and 2, echovirus types 1 and 5, and coxsackievirus types A9 and B5 were 99% inactivated in 0.3–4.5 min with residual chlorine levels of ~0.5 mg/l at pH values of 6.0 and 7.8; however, at pH 10, inactivation times were substantially increased (21–96 min) for all viruses except coxsackie A9, which was still rapidly inactivated after 1.5 min. Chlorine solutions at low pH (pH 6) were previously shown to inactivate enteric viruses more readily than chlorine solutions maintained at pH 10 [28]. Poliovirus was treated with 0.5 mg/l of free chlorine for various intervals, and virus propagation was attempted in cell culture followed by detection by reverse transcription polymerase chain reaction (RT-PCR) [17]. Poliovirus required 10 min for inactivation to non-detectable levels, demonstrating that poliovirus is five times more sensitive than previously expected [17].

Sattar *et al* [91] evaluated a host of potential disinfectants for the inactivation of rotaviruses and found that the best inactivation occurred with a 3-min exposure to a spray containing *o*-phenylphenol and ethanol; however, chlorine bleach and a phenolic compound were also effective in eliminating the viruses over a longer exposure period. A quarternary ammonium-based product was ineffective as a disinfectant [91].

Depending on the food type, chemical disinfection of viruses from surfaces of foods may be difficult or impossible to accomplish. Disinfectant solutions or detergents incorporated into the rinse water may facilitate the elimination of gross external contamination, but may be insufficient to remove viral contaminants, particularly if the viruses have penetrated through the skin. Washing and disinfection may not be effective for cut vegetables since viruses may enter the tissues through cuts and abrasions. Leafy vegetables may be more difficult to disinfect because of the rough or wrinkled nature of the surfaces. Likewise, fruits such as strawberries, raspberries, and blackberries have a porous surface texture which may trap viruses beyond the reach of chemical disinfectants. Rinsing without the use of suitable detergents or disinfectants may allow viruses to infiltrate the tissues of fruits and vegetables and to become redistributed onto products that were not previously contaminated. Therefore,

Table 1 Studies on the effectiveness of disinfectants for the reduction of enteric viruses

Enteric virus	Substrate	Reference
Coxsackie B3, adenovirus, coronavirus	Organic suspensions on stainless steel surface	Sattar *et al* [92]
Coxsackie B3 and B5, poliovirus	Buffers	Jensen *et al* [48]
Poliovirus	Water	Blackmer *et al* [17]
Poliovirus	Blood and saline solution	Weber *et al* [106]
Poliovirus	Buffers	Alvarez and O'Brien [3]
Hepatitis A virus	Stainless steel surface	Mbithi *et al* [65]
Hepatitis A and rotavirus	Polystyrene surface, fomites, suspensions	Abad *et al* [2]
Rotavirus	Steel, glass and plastic surfaces, instrument soaks	Lloyd-Evans *et al* [60]
Rotavirus	Stainless steel surface	Sattar *et al* [91]
Rotavirus	Water and dried organic suspensions (feces, media, milk)	Ward *et al* [104]
Rotavirus	Buffer	Chen and Vaughn [22]; Vaughn *et al* [101]
Feline calicivirus	Suspensions and dried preparations	Doultree *et al* [27]
Norwalk virus	Drinking water	Keswick *et al* [50]
Six enteroviruses	Buffer	Engelbrecht *et al* [28]

individual washing of produce would be better than bulk washing to prevent cross-contamination. Potential disinfectants and cleaning agents must be scrutinized carefully to ensure that they are approved for use in food establishments or on the foods themselves.

Sanitation of equipment and food contact surfaces should employ the use of ample detergents and water to rinse away and dilute — if nothing else — the viruses associated with food residues or hand contamination. Large-scale food processing plants should consider the use of commercial sanitation stations utilizing hot disinfectant solutions and high-pressure water rinses for inactivation of microorganisms and removal of food residues from processing equipment and food contact surfaces.

Cooking/heating

Thorough cooking is one of the best and most practical methods to totally inactivate enteric viruses; however, the times and temperatures required vary with the composition of the food. High protein and fat content tends to enhance thermal stability of enteric viruses [34]. Enteric viruses were inactivated from hamburgers heated to an internal temperature of 60°C (rare), but only when the patties were rapidly cooled after cooking [97]. Studies performed by adding $1.0-1.9\times10^4$ poliovirus per gram of oysters followed by stewing, frying, baking, and steaming gave varying results, depending on the cooking method [26]. Thirteen percent of the viruses remained infective in fried oysters cooked to an internal temperature of 100°C (requiring 8 min); whereas baking to 90° (requiring 29 min), steaming to 97.3°C (requiring 30 min), and stewing to 75°C (requiring 8 min) left 12.7%, 7%, and 10% of the viruses remaining, respectively [26]. Another gastroenteritis virus, the astrovirus, was inactivated by heating to 60°C for 10 min [55]. Studies on heat inactivation of hepatitis A virus in skim milk, homogenized milk, and table cream demonstrated a 5-log reduction with exposure of 85°C for <0.5 min; whereas at 80°C, skim and homogenized milk required up to 0.68 min and cream required 1.24 min for similar reductions [15]. Their findings [15] supported the conclusion that higher fat content provides thermal stability for hepatitis A virus.

Irradiation

Ionizing radiation, UV light, and microwave energy are all effective in reducing enteric viruses in foods. Viral nucleic acids are fragmented in the presence of ionizing radiation. Ionizing radiation, in the form of gamma emissions, is becoming more generally accepted for food processing [4,31,32]. Depending on the product, irradiation levels as high as 10 kGy may be used. A petition currently before the U.S. Food and Drug Administration seeks to allow the use of 4.5 kGy for non-frozen and non-dry products and 10.0 kGy for frozen or dry products [11]. Enteric virus inactivation requires a higher dose of gamma irradiation than bacterial pathogens [32,72,77]. The level of irradiation required to eliminate 90% of the hepatitis A virus, poliovirus, and rotavirus in shellfish was $\leq$3.1 kGy [63], but 6.8 kGy was required to inactivate coxsackievirus type B2 from ground beef [98]. In-shell molluscan shellfish experience high mortalities and reduced shelf-life when subjected to the levels of irradiation necessary to eliminate the enteric viruses. When poliovirus was suspended in water, 90% of the viruses was inactivated with 1.92 kGy, but 3.35 kGy was required to inactivate viruses in wastewater sludges [49]. In many cases, 90% inactivation may not be sufficient to eliminate the threat of illness, since only a few virus particles may be sufficient to elicit illness.

Ionizing radiation has the ability to penetrate foods and kill the microbes within; however, the opposite is true for UV irradiation which only inactivates viruses in direct contact with the light. Germicidal lamps, operating at a wavelength around 254 nm, are effective in reducing viruses on the surfaces of foods. Rotavirus and poliovirus demonstrated similar rates of inactivation from UV; however, these viruses required three to four times the dose of UV necessary for *Escherichia coli* inactivation [21].

Microwave energy inactivates viruses primarily as a function of the heat produced. Poliovirus added to infant formula was eliminated by microwaving [53]; however, the amount of microwave heating required to inactivate enteric viruses will probably vary depending on product composition [15]. Epidemiological evidence that microwaving of foods inactivates hepatitis A virus was reported [71]. A major limitation of microwave processing is that it heats foods unevenly, often leaving cold spots where viruses may survive. Thoroughly stirring or frequently turning foods during microwave processing can reduce the effects of unbalanced heating and ensure greater levels of virus reduction.

Dessication/dehydration

Viruses subjected to drying are generally perceived as more susceptible to inactivation than viruses stored in moist environments. Viruses from raw sewage sludge and wastewater were inactivated more quickly during drying [18,45]. Several enteric viruses were inactivated faster on a variety of porous and non-porous surfaces at low relative humidity [1], although there is one report that the infectivity of hepatitis A virus is inversely proportional to the level of relative humidity [66]. Airborne survival of enteric viruses is enhanced under conditions of high relative humidity [47]. Freeze-drying reduced poliovirus in potato salad, beef and vegetables, beef pot roast, chicken with gravy, and salmon salad by three to four orders of magnitude [40].

Depuration and relaying

Molluscan shellfish may undergo relaying, a process where shellfish are removed from marginally contaminated waters (as determined by coliform levels) and replanted in waters classified by state health authorities as approved (reviewed in Ref. [84]). Once in clean waters, shellfish will purge at least some of their contaminants, provided that the shellfish are physiologically active and the water conditions, temperature, salinity, and dissolved oxygen are favorable. A similar purging process, known as depuration, is conducted commercially in tanks of clean seawater. The water is usually recirculated through a UV or ozone disinfection system which kills or inactivates any bacteria or viruses present in the water (reviewed in Refs. [84,85]. Relaying requires extended periods (often 10 days to 2 weeks or more), whereas depuration may be completed within 48–72 h. In either case, viruses are reduced in shellfish, but are not totally eliminated; both depuration and relaying are more efficient in eliminating potential bacterial pathogens like *E. coli* and *Salmonella* spp. than the enteric viruses. Depurated shellfish can cause enteric virus illness, either as a consequence of improper depuration controls or due to insufficiency in the process itself [5,37]. For these reasons, only lightly contaminated shellfish should be subjected to depuration; more heavily contaminated shellstock should be relayed for extended periods. Shellfish-related industries should

appreciate the benefits of depuration in reducing bacterial contaminants and intestinal sediment; however, it must be recognized that there are no guarantees that shellfish depurated according to current technologies will be virologically safe.

Improved handling techniques

Increased automation of processing lines can significantly reduce contact between product and hands. Robotic technology provides for rapid and efficient processing of foods with minimal opportunity for hand contamination. For operations where hand contact of the food is common, proper disinfection of hands, gloves, or frequent replacement of disposable gloves may be necessary to reduce the opportunity for product contamination. Simple processing techniques to minimize direct human contact with foods can further safeguard the food supply. Perhaps, the simplest and most practical example of this concept is to mix salads, coleslaw, and similar products with a spoon or fork rather than by hand. Another example is to use clean food processors to slice and dice vegetables or cheeses destined for raw consumption rather than cut them by hand. Clean bowls or tubs should be used for mixing rather than sinks which are virtually impossible to thoroughly disinfect. At least one outbreak of foodborne Norwalk-like illness was associated with mixing potato salad in a sink [78].

Self-service, buffet-style restaurants should take precautions to protect foods from contamination by consumers. Too often, serving utensils at salad bars are so small that they fall into the food trays and have to be "fished out" by the consumer, usually using their fingers. Long-handled spoons, forks, and tongs reduce the possibility of direct consumer contact with foods. Single-use containers for salad dressings and condiments eliminate customer handling and possible contamination that may occur in multi-use containers. Sneeze guards are commonly used and, in most cases, mandated around buffet tables to prevent accidental contamination of foods by customers or servers.

Personal hygiene

Poor personal hygiene remains one of the greatest stumbling blocks in food safety. Providing employee education is a key to improving personal hygiene. Thorough disinfection of hands is difficult, since most chemicals are too harsh for the skin at the concentrations required for virus inactivation [23]. The use of disposable gloves remains a practical and inexpensive alternative to repeated hand disinfection [80]. Automatic or foot-controlled faucets in rest rooms would also reduce the possibility of recontaminating hands after washing. Frequent disinfection of rest room doors and door handles could further retard the spread of viruses. Restrooms should be maintained in a sanitary condition since they have been linked to the spread of enteric virus illness [43]. Although gloves, aprons, and good hygienic practices should reduce the risk of virus transmission to foods, people occasionally forget or disregard safety practices. Signs placed in strategic locations throughout the facility can serve as a reminder of acceptable and unacceptable practices.

Immunizations

Vaccines are available for hepatitis A virus, but there is none currently available for the Norwalk-like viruses, astroviruses, or rotaviruses. The RotaShield vaccine (Wyeth–Lederle Vaccines and Pediatrics) for rotavirus was licensed in the United States for oral administration to infants [9,44], but was recently withdrawn from the market due to unforseen bowel obstructions occurring in children who had received the vaccine. Immunizations of individuals who routinely handle foods could reduce the incidence of foodborne illness; however, conventional wisdom dictates that, rather than target at-risk groups, more broad-based immunization programs designed around a childhood immunization strategy would provide a sustained reduction of hepatitis A within the population. States with a higher-than-average incidence of hepatitis A should consider implementation of immunization programs for children. Two hepatitis A vaccines are available in the United States for children 2 years old and older: the Havrix® (1992 — SmithKline Beecham) and the Vaqta™ (1996 — Merck) [6]. Progress is being made on the development of a vaccine against hepatitis E virus [109]; however, its use in the United States is not warranted at this time. If the global distribution of hepatitis E changes, as might be expected with an ever-widening influx of foreign travel and demand for imported foods, hepatitis E immunizations may become necessary. The magnitude of the Norwalk-like outbreaks in the United States demonstrates the need for an efficacious vaccine; however, there have been obstacles to the development of such a vaccine [30].

Monitoring tools

The detection of foodborne viruses can be a difficult task. Procedures are available to extract and concentrate viruses from the surfaces of produce and from meats and shellfish [87]; however, most extraction and assay procedures are time-consuming and of limited utility in routine monitoring programs. Traditional assay techniques rely on the propagation of "indicator" viruses since Norwalk-like viruses cannot be propagated in cell cultures, and hepatitis A virus is very difficult to assay. For many years, it was possible to test for the presence of poliovirus in foods since the vaccine strains, commonly shed in the stools of recently immunized children, were indicative of the presence of fecal pollution of human origin and of the possible presence of hepatitis A, Norwalk, and other human enteric viruses for which tests were not available. The efforts of the World Health Assembly have been highly effective in eradicating poliomyelitis from most of the world. The recent elimination of the use of oral polio vaccines in some countries has reduced poliovirus levels in the environment and negated the use of poliovirus as a potential indicator organism.

The advent of molecular biology has led to a plethora of techniques to assay viruses; however, the extraction of viruses from the foods remains a difficult task [87]. Molecular techniques are excellent for epidemiological investigations and are amenable for detecting a wide variety of viruses including hepatitis A and E, the caliciviruses, and astroviruses. Unfortunately, molecular techniques have limited usefulness in predicting the virological safety of foods [86]. Molecular techniques generally involve RT-PCR to detect hepatitis A and Norwalk-like viruses; however, the procedure cannot differentiate between infectious and inactivated viruses, and is unable, under most circumstances, to provide a quantitative measure of the level of virus present [86].

New, innovative approaches are needed to identify the presence of infectious viruses in water and foods. Improved cell culture techniques are needed to propagate wild-type hepatitis A and Norwalk-like viruses. Currently, cell-culture-adapted strains of hepatitis A virus are available for laboratory studies and a feline calicivirus model is available for studies of Norwalk-like viruses. With additional studies on virus–receptor interactions and the use of alternative cell cultures or treatments, it may become possible in the future to directly monitor for hepatitis A and E viruses, caliciviruses, astroviruses, and other viral pathogens in foods.

References

1 Abad FX, RM Pinto and A Bosch. 1994. Survival of enteric viruses on environmental fomites. *Appl Environ Microbiol* 60: 3704–3710.
2 Abad FX, RM Pinto and A Bosch. 1997. Disinfection of human enteric viruses on fomites. *FEMS Microbiol Lett* 156: 107–111.
3 Alvarez ME and RT O'Brien. 1982. Mechanisms of inactivation of poliovirus by chlorine dioxide and iodine. *Appl Environ Microbiol* 44: 1064–1071.
4 Andrews LS, M Ahmedna, RM Grodner, JA Liuzzo, PS Murano, EA Murano, RM Rao, S Shane and PW Wilson. 1998. Food preservation using ionizing radiation. *Rev Environ Contam Toxicol* 154: 1–53.
5 Ang LH. 1998. An outbreak of viral gastroenteritis associated with eating raw oysters. *Commun Dis Public Health* 1: 38–40.
6 Anonymous. 1996. Prevention of hepatitis A infections: guidelines for use of hepatitis A vaccine and immune globulin. American Academy of Pediatrics Committee on Infectious Diseases. *Pediatrics* 98: 1207–1215.
7 Anonymous. 1997a. Hepatitis A associated with consumption of frozen strawberries — Michigan, March 1997. *Morb Mortal Wkly Rep* 46: 288, 295.
8 Anonymous. 1997b. Viral gastroenteritis associated with eating oysters — Louisiana, December 1996–January 1997. *Morb Mortal Wkly Rep* 46: 1109–1112.
9 Anonymous. 1998. Prevention of rotavirus disease: guidelines for use of rotavirus vaccine. American Academy of Pediatrics. *Pediatrics* 102: 1483–1491.
10 Anonymous. 1999. Food safety issues associated with products from aquaculture. Report of a Joint FAO/NACA/WHO Study Group. *World Health Organ Tech Rep Ser* 883: 1–55.
11 Anonymous. 2000. FDA extends review on ready-to-eat food irradiation petition for 90 more days. *Food Chem News* 42: 10–11.
12 Beller N. 1992. Hepatitis A outbreak in Anchorage, Alaska, traced to ice slush beverages. *West J Med* 156: 624–627.
13 Berg DE, MA Kohn, TA Farley and LM McFarland. 2000. Multi-state outbreaks of acute gastroenteritis traced to fecal-contaminated oysters harvested in Louisiana. *J Infect Dis* 181: S381–S386.
14 Beuchat RL and JH Ryu. 1997. Produce handling and processing practices. *Emerging Infect Dis* 3: 459–465.
15 Bidawid S, JM Farber, SA Satter and S Hayward. 2000. Heat inactivation of hepatitis A virus in dairy foods. *J Food Prot* 63: 522–528.
16 Billy TJ and IK Wachsmuth. 1997. Hazard analysis and critical control point systems in the United States Department of Agriculture regulatory policy. *Rev Sci Technol* 16: 342–348.
17 Blackmer F, KA Reynolds, CP Gerba and IL Pepper. 2000. Use of integrated cell culture PCR to evaluate the effectiveness of poliovirus inactivation by chlorine. *Appl Environ Microbiol* 66: 2267–2268.
18 Brachear DA and RL Ward. 1983. Inactivation of indigenous viruses in raw sludge by air drying. *Appl Environ Microbiol* 45: 1943–1945.
19 Bradley DW. 1992. Hepatitis E: epidemiology, aetiology and molecular biology. *Rev Med Virol* 2: 19–28.
20 Burkhardt W III and KR Calci. 2000. Selective accumulation may account for shellfish-associated viral illness. *Appl Environ Microbiol* 66: 1375–1378.
21 Chang JCH, SF Ossoff, DC Lobe, MH Dorfman, CM Dumais, RG Qualls and JD Johnson. 1985. UV inactivation of pathogenic and indicator microorganisms. *Appl Environ Microbiol* 49: 1361–1365.
22 Chen YS and JM Vaughn. 1990. Inactivation of human and simian rotaviruses by chlorine dioxide. *Appl Environ Microbiol* 56: 1363–1366.
23 Cliver DO and KD Kostenbader Jr. 1984. Disinfection of virus on hands for prevention of foodborne disease. *Int J Food Microbiol* 1: 75–87.
24 Daniels NA, DA Bergmire-Sweat, KJ Schwab, KA Hendricks, S Reddy, SM Rowe, RL Fankhausser, SS Monroe, RL Atmar, RI Glass and P Mead. 2000. A foodborne outbreak of gastroenteritis associated with Norwalk-like viruses: first molecular traceback to deli sandwiches contaminated during preparation. *J Infect Dis* 181: 1467–1470.
25 Deng MY and DO Cliver. 1995. Persistence of inoculated hepatitis A virus in mixed human and animal wastes. *Appl Environ Microbiol* 61: 87–91.
26 DiGirolamo R, J Liston and JR Marches. 1970. Survival of virus in chilled, frozen and processed oysters. *Appl Microbiol* 20: 58–63.
27 Doultree JC, JD Druce, CJ Birch, DS Bowden and JA Marshall. 1999. Inactivation of feline calicivirus, a Norwalk virus surrogate. *J Hosp Infect* 41: 51–57.
28 Engelbrecht RS, MJ Weber, BL Salter and CA Schmidt. 1980. Comparative inactivation of viruses by chlorine. *Appl Environ Microbiol* 40: 249–256.
29 Erker JC, SM Desai, GC Schlauder, GJ Dawson and IK Mushahwar. 1999. A hepatitis E variant from the United States: molecular characterization and transmission in cynomolgus macaques. *J Gen Virol* 80: 681–690.
30 Estes MK, JM Ball, RA Guerrero, AR Opekum, MA Gilger, SS Pacheco and DY Graham. 2000. Norwalk virus vaccines: challenges and progress. *J Infect Dis* 181: S367–S373.
31 Farkas J. 1998. Irradiation as a method of decontaminating food. A review. *Int J Food Microbiol* 44: 189–204.
32 Farkas J. 1989. Microbiological safety of irradiated foods. A review. *Int J Food Microbiol* 9: 1–15.
33 Fattal B, M Margalith, HI Shuval, Y Wax and A Morag. 1987. Viral antibodies in agricultural populations exposed to aerosols from wastewater irrigation during a viral disease outbreak. *Am J Epidemiol* 125: 899–906.
34 Filippi JA and GJ Banwart. 1974. Effect of the fat content of ground beef on the heat inactivation of poliovirus. *J Food Sci* 39: 865–868.
35 Gaulin CD, D Ramsay, P Cardinal and MA D'Halevyn. 1999. Epidemic of gastroenteritis of viral origin associated with eating imported raspberries. *Can J Public Health* 90: 37–40.
36 Griffin MR, JJ Surowiec, DI McCloskey, B Capuano, B Pierzynski, M Quinn, R Wojnarski, WE Parkin, H Greenberg and GW Gary. 1982. Foodborne Norwalk virus. *Am J Epidemiol* 115: 178–184.
37 Grohmann GS, AM Murphy, PJ Christopher, E Auty and HB Greenberg. 1981. Norwalk virus gastroenteritis in volunteers consuming depurated oysters. *Aust J Exp Biol Med Sci* 59: 219–228.
38 Halliday ML, LY Kang, TK Zhou, MD Hu, QC Pan, TY Fu, YS Huang and SL Hu. 1991. An epidemic of hepatitis A attributable to the ingestion of raw clams in Shanghai, China. *J Infect Dis* 164: 852–859.
39 Harakeh M and M Butler. 1984. Inactivation of human rotavirus SA11 and other enteric viruses in effluent by disinfectants. *J Hyg (Cambridge)* 93: 157–163.
40 Heidelbaugh ND and DJ Giron. 1969. Effects of processing on recovery of polio virus from inoculated foods. *J Food Sci* 34: 239–241.
41 Hernandez F, R Monge, C Jimenez and L Taylor. 1997. Rotavirus and hepatitis A virus in market lettuce. *Int J Food Microbiol* 37: 221–223.
42 Herwaldt BL, JF Lew, CL Moe, DC Lewis, CD Humphrey, SS Monoe, EW Pon and RI Glass. 1994. Characterization of a variant strain of Norwalk virus from a foodborne outbreak of gastroenteritis on a cruise ship in Hawaii. *J Clin Microbiol* 32: 861–866.
43 Ho MS, RI Glass, SS Monroe, HP Madore, S Stine, PF Pinsky, D Cubitt, C Ashley and EO Caul. 1989. Viral gastroenteritis aboard a cruise ship. *Lancet* 2 (8669): 961–965.
44 Hochwald C and L Kivela. 1999. Rotavirus vaccine, live, oral, tetravalent. *Pediatr Nurs* 25: 203, 207.
45 Hurst CJ, CP Gerba, JC Lance and RC Rice. 1980. Survival of enteroviruses in rapid-infiltration basins during the land application of wastewater. *Appl Environ Microbiol* 40: 192–200.
46 Hutin YJ, V Pool, EH Cramer, OV Nainan, J Weth, IT Williams, ST Goldstein, KF Gensheimer, BP Bell, CN Shapiro, MJ Alter and HS

Margolis. 1999. A multistate, foodborne outbreak of hepatitis A. National Hepatitis A Investigation Team. *N Engl J Med* 340: 595–602.

47 Ijaz MK, SA Sattar, CM Johnson-Lussenburg and VS Springthorpe. 1985. Comparison of the airborne survival of calf rotavirus and poliovirus type 1 (Sabin) aerosolized as a mixture. *Appl Environ Microbiol* 49: 289–293.

48 Jensen H, K Thomas, DG Sharp. 1980. Inactivation of coxsackie viruses B3 and B5 in water by chlorine. *Appl Environ Microbiol* 40: 633–640.

49 Kaupert N, E Burgi and L Scolaro. 1999. Inactivation of poliovirus by gamma irradiation of wastewater sludges. *Rev Argent Microbiol* 31: 49–52.

50 Keswick BH, TK Satterwhite, PC Johnson, HL Dupont, SL Secor, JA Bitsura, GW Gary and JC Hoff. 1985. Inactivation of Norwalk virus in drinking water by chlorine. *Appl Environ Microbiol* 50: 261–264.

51 Khan AS, CL Moe, RI Glass, SS Monroe, MK Estes, LE Chapman, X Jiang, C Humphrey, E Pon and JK Iskander. 1994. Norwalk virus-associated gastroenteritis traced to ice consumption aboard a cruise ship in Hawaii: comparison and application of molecular method-based assays. *J Clin Microbiol* 32: 318–322.

52 Kilgore PE, ED Belay, DM Hamlin, JS Noel, CD Humphrey, HE Gary Jr, T Ando, SS Monroe, PE Kludt, DS Rosenthal, J Freeman and RI Glass. 1996. A university outbreak of gastroenteritis due to a small round-structured virus. Application of molecular diagnostics to identify the etiologic agent and patterns of transmission. *J Infect Dis* 173: 787–793.

53 Kindle G, A Busse, D Kampa, U Meyer-Konig and FD Daschner. 1986. Killing activity of microwaves in milk. *J Hosp Infect* 33: 273–278.

54 Kohn MA, TA Farley, T Ando, M Curtis, SA Wilson, Q Jin, SS Monroe, RC Baron, LM McFarland and RI Glass. 1995. An outbreak of Norwalk virus gastroenteritis associated with eating raw oysters. Implications for maintaining safe oyster beds. *J Am Med Assoc* 273: 466–471.

55 Kurtz JB and TW Lee. 1987. Astroviruses: human and animal. *Ciba Found Symp* 128: 92–107.

56 Kwo PY, GG Schlauder, HA Carpenter, PJ Murphy, JE Rosenblatt, GJ Dawson, EE Mast, K Krawczynski and V Balan. 1997. Acute hepatitis E by a new isolate acquired in the United States. *Mayo Clin Proc* 72: 1133–1136.

57 Lemon SM and CN Shapiro. 1994. The value of immunization against hepatitis A. *Infect Agents Dis* 3: 38–49.

58 Levine WC, WT Stephenson and GF Craun. 1990. Waterborne disease outbreaks, 1986–1988. *Morb Mortal Wkly Rep CDC Surveill Summ* 39: 1–13.

59 Linnemann CC Jr, R Jaffa, PS Gartside, PV Scarpino and CS Clark. 1984. Risk of infection associated with a wastewater spray irrigation system used for farming. *J Occup Med* 26: 41–44.

60 Lloyd-Evans N, VS Springthorp and SA Sattar. 1986. Chemical disinfection of human rotavirus-contaminated inanimate surfaces. *J Hyg* 97: 163–173.

61 Lo SV, AM Connolly, SR Palmer, D Wright, PD Thomas and D Joynson. 1994. The role of the pre-symptomatic food handler in a common source outbreak of foodborne SRSV gastroenteritis in a group of hospitals. *Epidemiol Infect* 113: 513–521.

62 Lowry PW, R Levine, DF Stroup, RA Gunn, MH Wilder and C Konigsberg Jr. 1989. Hepatitis A outbreak on a floating restaurant in Florida, 1986. *Am J Epidemiol* 129: 155–164.

63 Mallet JC, LE Beghian, TG Metcalf and JD Kaylor. 1991. Potential of irradiation technology for improving shellfish sanitation. *J Food Saf* 11: 231–245.

64 Mast EE and K Krawczynski. 1996. Hepatitis E: an overview. *Annu Rev Med* 47: 257–266.

65 Mbithi JN, VS Springthorpe and SA Sattar. 1990. Chemical disinfection of hepatitis A virus on environmental surfaces. *Appl Environ Microbial* 56: 3601–3604.

66 Mbithi JN, VS Springthorpe and SA Sattar. 1991. Effect of relative humidity and air temperature on survival of hepatitis A virus on environmental surfaces. *Appl Environ Microbiol* 57: 1394–1399.

67 McDonnell S, KB Kirkland, WG Hlady, C Aristeguieta, RS Hopkins, SS Monroe and RI Glass. 1997. Failure of cooking to prevent shellfish-associated viral gastroenteritis. *Arch Intern Med* 157: 111–116.

68 McDonnell RJ, PG Wall, GK Adak, HS Evans, JM Cowden and EO Caul. 1995. Outbreaks of infectious intestinal disease associated with person-to-person spread in hotels and restaurants. *Commun Dis Rep CDR Rev* 15: R150–R152.

69 McEvoy M, W Blake, D Brown, J Green and R Cartwright. 1996. An outbreak of viral gastroenteritis on a cruise ship. *Commun Dis Rep CDR Rev* 6: R188–192.

70 Mead PS, L Slutsker, V Dietz, LF McCaig, JS Bresee, C Shapiro, PM Griffin and RV Tauxe. 1999. Food-related illness and death in the United States. *Emerging Infect Dis* 5: 607–625.

71 Mishu B, SC Hadler, VA Boaz, RH Hutcheson, JM Horan and W Schaffner. 1990. Foodborne hepatitis A: evidence that microwaving reduces risk. *J Infect Dis* 162: 655–658.

72 Monk JD, LR Beuchat and MP Doyle. 1995. Irradiation inactivation of foodborne microorganisms. *J Food Prot* 58: 197–208.

73 Morens DM, RM Zweighaft, TM Vernon, GW Gary, JJ Eslien, BT Wood, RC Holman and R Dolin. 1979. A waterborne outbreak of gastroenteritis with secondary person-to-person spread. Associations with a viral agent. *Lancet* 1 (8123): 964–966.

74 Niu MT, LB Polish, BH Roberston, BK Khanna, BA Woodruff, CN Shapiro, MA Miller, JD Smith and JK Gedrose. 1992. Multistate outbreak of hepatitis A associated with frozen strawberries. *J Infect Dis* 166: 518–524.

75 Oron G, M Goemans and Y Manor. 1995. Poliovirus distribution in the soil–plant system under reuse of secondary wastewater. *Water Res* 29: 1069–1078.

76 Parashar UD, JS Bresse, JR Gentsch and RI Glass. 1998. Rotavirus. *Emerging Infect Dis* 4: 561–570.

77 Patterson MF. 1993. Food irradiation and food safety. *Rev Med Microbiol* 4: 151–158.

78 Patterson W, P Haswell, PT Fryers and J Green. 1997a. Outbreak of small round-structured virus gastroenteritis after kitchen worker vomited. *Commun Dis Rep CDR Rev* 7: R101–103.

79 Patterson T, P Hutchings and S Palmer. 1997b. Outbreak of SRSV gastroenteritis at an international conference traced to food handling by a post-symptomatic caterer. *Epidemiol Infect* 111: 157–162.

80 Paulson DS. 1997. Foodborne disease: controlling the problem. *J Environ Health* 59: 15–19.

81 Powell SC and R Attwell. 1999. The use of epidemiological data in the control of foodborne viruses. *Rev Environ Health* 14: 31–37.

82 Reid TM and HG Robinson. 1987. Frozen raspberries and hepatitis A. *Epidemiol Infect* 98: 109–112.

83 Richards GP. 1985. Outbreaks of shellfish-associated enteric virus illness in the United States: requisite for development of viral guidelines. *J Food Prot* 48: 815–823.

84 Richards GP. 1988. Microbial purification of shellfish: a review of depuration and relaying. *J Food Prot* 51: 218–251.

85 Richards GP. 1990. Shellfish depuration. In: Ward DR and Hackney CR (Eds.), Microbiology of Marine Food Products. Van Nostrand-Reinhold, New York, NY, pp. 395–428.

86 Richards GP. 1999. Limitations of molecular biological techniques for assessing the virological safety of foods. *J Food Prot* 62: 691–697.

87 Richards GP and DO Cliver. Foodborne viruses. In: Compendium of Methods for the Microbiological Examination of Foods. American Public Health Assoc., Washington, DC, in press.

88 Robertson BH and SM Lemon. 1998. Hepatitis A and E. In: Collier C, Ballows A and Sussman M (Eds.), Topley and Wilson's Microbiology and Microbial Infections. Virology. Oxford University Press, New York, NY, pp. 693–716.

89 Rosenblum LS, IR Mirkin, DT Allen, S Safford and SC Hadler. 1990. A multifocal outbreak of hepatitis A traced to commercially distributed lettuce. *Am J Public Health* 80: 1075–1079.

90 Sadovski AY, B Fattal, D Goldberg, E Katzenelson and HI Shuval. 1978. High levels of microbial contamination of vegetables irrigated with wastewater by the drip method. *Appl Environ Microbiol* 36: 824–830.

91 Sattar SA, H Jacobsen, H Rahman, TM Cusack and JR Rubino. 1994. Interruption of rotavirus spread through chemical disinfection. *Infect Control Hosp Epidemiol* 15: 751–756.

92 Sattar SA, VS Springthorpe, Y Karim and P Loro. 1989. Chemical disinfection of non-porous inanimate surfaces experimentally contaminated with four human pathogenic viruses. *Epidemiol Infect* 102: 493–505.

93 Slomka MJ and H Appleton. 1998. Feline calicivirus as a model system for heat inactivation studies of small round-structured viruses in shellfish. *Epidemiol Infect* 121: 401–407.
94 Sobsey MD, PA Shields, FS Hauchman, AL Davis, VA Rullman and A Bosch. 1988. Survival and persistence of hepatitis A virus in environmental samples. In: Zucherman AJ (Ed.), Viral Hepatitis and Liver Disease. Alan R. Liss, New York, pp. 121–124.
95 Spillmann SK, F Traub, M Schwyzer and R Wyler. 1987. Inactivation of animal viruses during sewage sludge treatment. *Appl Environ Microbiol* 53: 2077–2081.
96 Straub TM, IL Pepper and CP Gerba. 1993. Hazards from pathogenic microorganisms in land-disposed sewage sludge. *Rev Environ Contam Toxicol* 132: 55–91.
97 Sullivan R, RM Marnell, EP Larkin and RB Read Jr. 1975. Inactivation of poliovirus 1 and coxsackievirus B-2 in broiled hamburgers. *J Milk Food Technol* 38: 473–475.
98 Sullivan R, PV Scarpino, AC Fassolitis, EP Larkin and JT Peeler. 1973. Gamma radiation inactivation of coxsackievirus B-2. *Appl Microbiol* 26: 14–17.
99 Taylor JW, GW Gary Jr, HB Greenberg. 1981. Norwalk-related viral gastroenteritis due to contaminated drinking water. *Am J Epidemiol* 114: 584–592.
100 U.S. Department of Health and Human Services. 1997. National Shellfish Sanitation Program Guide for the Control of Molluscan Shellfish (Revision). Shellfish Program Implementation Branch, U.S. Food and Drug Administration, Washington, DC.
101 Vaughn JM, YS Chen, K Lindburg and D Morales. 1987. Inactivation of human and simian rotaviruses by ozone. *Appl Environ Microbiol* 53: 2218–2221.
102 Vaughn JM, YS Chen and MZ Thomas. 1986. Inactivation of human and simian rotaviruses by chlorine. *Appl Environ Microbiol* 51: 391–394.
103 Warburton AR, TG Wreghitt, A Rampling, R Buttery, KN Ward, KR Perry and JV Parry. 1991. Hepatitis A outbreak involving bread. *Epidemiol Infect* 106: 199–202.
104 Ward RL, DI Bernstein, DR Knowlton, JR Sherwood, EC Young, TM Cusack, JR Rubino and GM Schiff. 1991. Prevention of surface-to-human transmission of rotaviruses, by treatment with disinfectant spray. *J Clin Microbiol* 29: 1991–1996.
105 Warner RD, RW Carr, FK McCleskey, PC Johnson, LM Elmer and VE Davison. 1991. A large nontypical outbreak of Norwalk virus. Gastroenteritis associated with exposing celery to non-potable water and with *Citrobacter frundii*. *Arch Intern Med* 151: 2419–2424.
106 Weber DJ, SL Barbee, MD Sobsey and WA Rutala. 1999. The effect of blood on the antiviral activity of sodium hypochlorite, a phenolic, and a quaternary ammonium compound. *Infect Control Hosp Epidemiol* 20: 821–827.
107 Weltman AC, NM Bennett, DA Ackman, JH Misage, JJ Campana, LS Fine, AS Doniger, GJ Balzano and GS Birkhead. 1996. An outbreak of hepatitis A associated with a bakery, New York, 1994: the 1968 "West Branch, Michigan" outbreak repeated. *Epidemiol Infect* 117: 333–341.
108 White KE, MT Osterholm, JA Mariotti, JA Korlath, DH Lawrence, TL Ristinen and HB Greenberg. 1996. A foodborne outbreak of Norwalk virus gastroenteritis: evidence for post-recovery transmission. *Am J Epidemiol* 124: 120–126.
109 Yarbough PO. 1999. Hepatitis E virus. Advances in HEV biology and HEV vaccine approaches. *Intervirology* 42: 179–184.