Introduction

Raw fruits and vegetables have been known to serve as vehicles of human disease for at least a century. In 1899, Morse linked typhoid infection to eating celery. Warry (1903) attributed an outbreak of typhoid fever to eating watercress grown in soil fertilized with sewage and Pixley (1913) recorded two cases of typhoid from eating uncooked rhubarb which was grown in soil known to have been fertilized with typhoid excreta. In 1912, Creel demonstrated that lettuce and radishes grown in soil containing *Bacillus typhosa* (now *Salmonella* Typhi) harboured the organism on their surfaces for up to 31 days. Melick (1917) recovered typhoid bacilli from mature lettuce and radish harvested from soil that had been inoculated at the time seeds were planted. Some parasitic helminths (e.g. *Fasciola hepatica*, *Fasciolopsis buski*) require encystment on plants to complete their life cycle. Thus, the recognition of raw fruits and vegetables as potential vehicles for transmission of pathogenic microorganisms known to cause human disease is not new. Nevertheless, documented outbreaks of foodborne illness associated with fruits and vegetables in industrialized countries are relatively rare. For instance, in 1996 only six of about 200 reported foodborne disease outbreaks in the United Kingdom were associated with consumption of fruits and vegetables (PHLS, 1996). In recent years, however, the frequency of outbreaks epidemiologically associated with raw fruits and vegetables is documented to have increased in some industrialized countries (e.g the United States) as a result of change in dietary habits and increased import of food (Altekruse *et al.*, 1997). In developing countries, foodborne illnesses caused by contaminated fruits and vegetables are frequent and in some areas they cause a large proportion of illness. However, due to lack of foodborne disease investigation and surveillance in most of these countries, most outbreaks go undetected and the scientific literature reports only on very few outbreaks. In 1995-1996, only 2% of foodborne disease outbreaks in Latin America were related to fruits and vegetables (PAHO/INPPAZ, 1996).

Raw and minimally-processed fruits and vegetables are an essential part of people's diet all around the world. Where land is available, families grow fruits and vegetables for their own use. Alternatively, produce is purchased from local farmers or retail outlets for further preparation by street vendors, by families at home or as part of meals eaten in restaurants and other food-service facilities. While advances in agronomic practices, processing, preservation, distribution and marketing have enabled the raw fruit and vegetable industry to supply high-quality produce to many consumers all year round, some of these same practices have also expanded the geographical distribution and incidence of human illness associated with an increasing number of pathogenic bacterial, viral and parasitic microorganisms.

Changes that may contribute to the increase in diseases associated with the consumption of raw fruits and vegetables in industrialized countries (Hedberg *et al.*, 1994) and foods in general (Altekruse and Swerdlow, 1996; Altekruse *el al.*, 1997; Potter *et al.*, 1997) have been described. Factors include globalization of the food supply, inadvertent introduction of pathogens into new geographical areas — e.g. outbreaks of shigellosis in Norway, Sweden and the United Kingdom in 1994 due to contaminated lettuce imported from southern Europe (Frost et al., 1995; Kapperud et al., 1995) and cyclosporiasis in the United States which was linked to consumption of contaminated raspberries imported from Guatemala (Centers for Disease Control and Prevention, 1996c) — and the development of new virulence factors by microorganisms, decreases in immunity among certain segments of the population, and changes in eating habits. In developing countries, continued use of untreated wastewater and manure as fertilizers for the production of fruits and vegetables is a major contributing factor to contamination that causes numerous foodborne disease outbreaks.

While every effort should be made to prevent contamination of fruits and vegetables during production, transport, processing and handling, much improvement is still needed in some parts of the world if hygienic production of fruits and vegetables is to be ensured. Furthermore, many microbial contaminants are part of the environment and fruits and vegetables may be inadvertently contaminated. Unless measures are taken to decontaminate them, their safety may not be assured. This document provides an overview of the hazards associated with fruits and vegetables eaten raw, reviews the methods used for their decontamination (particularly with reference to chemical methods) and evaluates, on the basis of available scientific data, the efficacy of these methods. The purpose of this review is to provide public health authorities with information on the surface decontamination of fruits and vegetables eaten raw, and with guidelines and recommendations that can be provided to growers, processors and consumers.

Recognizing the problem

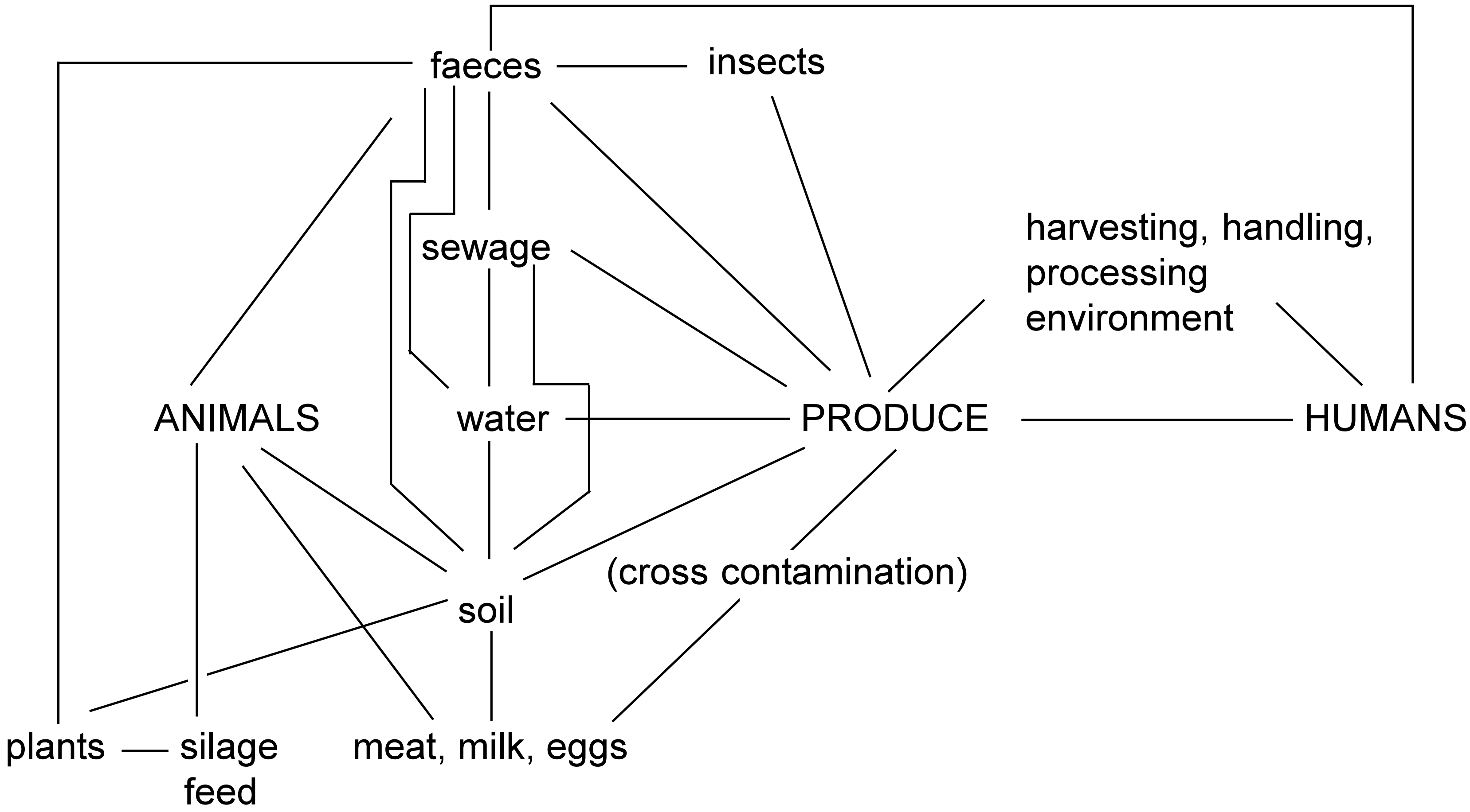
Fruits and vegetables can become contaminated with microorganisms capable of causing human diseases while still on the plant in fields or orchards, or during harvesting, transport, processing, distribution and marketing, or in the home. Figure 1 shows potential mechanisms by which pathogenic microorganisms can contaminate vegetables. Bacteria such as *Clostridium botulinum, Bacillus cereus* and *Listeria monocytogenes*, all capable of causing illness, are normal inhabitants of many soils, whereas *Salmonella, Shigella, Escherichia coli* and *Campylobacter* reside in the intestinal tracts of animals, including humans, and are more likely to contaminate raw fruits and vegetables through contact with faeces, sewage, untreated irrigation water or surface water. Contamination may also occur during post-harvest handling, including at points of preparation by street vendors, in food-service establishments and in the home. Contamination with viruses or parasites can result from contact with faeces, sewage and irrigation water (Cliver, 1997; Speer, 1997).

Numerous surveys have been carried out in many countries to determine the presence of pathogenic microorganisms on raw fruits and vegetables. In many instances, bacterial pathogens have not been detected. In other investigations, high percentages of samples have been found to contain bacteria capable of causing human disease. Results of these investigations are summarized in Table 1. This list is not comprehensive but it reflects the scope of potential problems that may link raw fruits and vegetables to human disease. Indeed, numerous outbreaks of illness caused by bacteria, viruses and parasites have been linked epidemiologically to consumption of raw fruits and vegetables (Table 2). Increased recognition of raw fruits and vegetables as suspected vehicles of human illness in industrialized countries (Altekruse *et al.*, 1997; Bean and Griffin, 1990; Bean *et al.*, 1997; Centers for Disease Control and Prevention, 1996b) may probably result in increased numbers of associated or confirmed cases in the future. Improved diagnostic systems and surveillance programmes will enhance the prospect of identifying raw fruits and vegetables as sources of foodborne illness, thus possibly resulting in an increase in recorded numbers of outbreaks and cases of disease. Surveys of raw fruits and vegetables for the presence of parasites or viruses are few, largely because of the lack of sensitive methods for their detection in plant materials.

Other factors, however, may contribute to real increases in diseases associated with fruits and vegetables (Hedberg *et al.*, 1994). These include use of wastewater, increased application of improperly composted manures to soils in which fruits and vegetables are grown, changes in packaging technology such as the use of modified or controlled atmosphere and vacuum packaging, extended time between harvesting and consumption, and changing food consumption patterns (e.g. eating more meals away from home, including greater use of salad bars). Increased global trade in raw fruits and vegetables, as well as increased international travel in general, could also increase the risk of produce-associated diseases. Finally, the susceptibility of the public to foodborne diseases, at least in more developed countries, is changing due to increased numbers of people who are elderly, immunocompromised or have chronic diseases. This change in social demographics is likely to lead to increased risk of illness associated with the consumption of raw produce that otherwise may contain levels of pathogens innocuous to healthy individuals.

Prevention of contamination of fruits and vegetables with pathogenic microorganisms should be the goal of everyone involved in both the pre-harvest and post-harvest phases of delivering produce to the consumer. This is a very difficult task, since some pathogens are normally present in the soil and may therefore be present on the surface of fruits and vegetables when they are harvested. In

Figure 1. Mechanisms by which raw fruits and vegetables may become contaminated with pathogenic microorganisms.



From Beuchat (1996b), and used with permission from the International Association of Milk, Food and Environmental Sanitarians.

Table 1. Bacterial pathogens isolated from raw vegetables[1]

Vegetable	Country	Pathogen	Prevalence	Reference
Alfalfa	United States	*Aeromonas*		Callister and Agger (1989)
Artichoke	Spain	*Salmonella*	3/25 (12.0%)	Garcia-Villanova Ruiz *et al.* (1987b)
Asparagus	United States	*Aeromonas*		Berrang *et al.*
Bean sprouts	Malaysia	*L. monocytogenes*	6/7 (85%)	Arumugaswamy *et al.* (1994)
	Malaysia	*Salmonella*	2/10 (20%)	Arumugaswamy *et al.* (1994)
	Sweden	*Salmonella*		Andersson and Jong (1989)
	Thailand	*Salmonella*	30/344 (8.7%)	Jerngklinchan and Saitanu (1993)
Beet leaves	Spain	*Salmonella*	4/52 (7.7%)	Garcia-Villanova Ruiz *et al.* (1987b)
Broccoli	Canada	*L. monocytogenes*	2/15 (13.3%)	Odumeru *et al.* (1997)
	United States	*Aeromonas*		Berrang *et al.* (1989)
	United States	*Aeromonas*	5/16 (31.3%)	Callister and Agger (1989)
Cabbage	Canada	*L. monocytogenes*	2/92 (2.2%)	Schlech *et al.* (1983)
	Canada	*L. monocytogenes*	1/15 (6.7%)	Odumeru *et al.* (1997)
	Mexico	*E. coli* O157:H7	1/4 (25.0%)	Zepeda-Lopez *et al.* (1995)
	Peru	*V. chlolerae*		Swerdlow *et al.* (1992)
	Saudi Arabia	*L. monocytogenes*		Salamah (1993)
	Saudi Arabia	*Y. enterocolitica*		Salamah (1993)
	Spain	*Salmonella*	7/41 (17.1%)	Garcia-VillanovaRuiz *et al.*(1987b)
	United States	*C. botulinum*	1/337 (0.3%)	Lilly *et al.* (1996)
	United States	*L. monocytogenes*	1/92 (1.1%)	Heisick *et al.* (1989b)
Carrot	Lebanon	*Staphylococcus*	(14.3%)	Abdelnoor *et al.* (1983)
	Saudi Arabia	*L. monocytogenes*		Salamah (1993)
	Saudi Arabia	*Y. enterocolitica*		Salamah (1993)
Cauliflower	Netherlands	*Salmonella*	1/13 (7.7%)	Tamminga *et al.* (1978)
	Spain	*Salmonella*	1/23 (4.5%)	Garcia-Villonova Ruiz*etal.* (1987b)
	United States	*Aeromonas*		Berrang *et al.* (1989)
Celery	Mexico	*E. coli* O157:H7	6/34 (17.6%)	Zepeda-Lopez *et al.* (1995)
	Spain	*Salmonella*	2/26 (7.7%)	Garcia-Villanova Ruiz*etal.*(1987b)
Chili	Surinam	*Salmonella*	5/16 (31.3%)	Tamminga *et al.* (1978)
Cilantro	Mexico	*E. coli* O157:H7	8/41 (19.5%)	Zepeda-Lopez *et al.* (1995)
Coriander	Mexico	*E. coli* O157:H7	2/10 (20.0%)	Zepeda-Lopez *et al.* (1995)
Cress sprouts	United States	*B. cereus*		Portnoy *et al.* (1976)
Cucumber	Malaysia	*L. monocytogenes*	4/5 (80%)	Arumugaswamy *et al.* (1994)
	Pakistan	*L. monocytogenes*	1/15 (6.7%)	Vahidy (1992)
	Saudi Arabia	*L. monocytogenes*		Salamah (1993)
	Saudi Arabia	*Y. enterocolitica*		Salamah (1993)
	United States	*L. monocytogenes*		Heisick *et al.* (1989b)
Egg plant	Netherlands	*Salmonella*	2/13 (1.5%)	Tamminga *et al.* (1978)
Endive	Netherlands	*Salmonella*	2/26 (7.7%)	Tamminga *et al.* (1978)
Fennel	Italy	*Salmonella*	4/89 (71.9%)	Ercolani (1976)
Green onion	Canada	*Campylobacter*	1/40 (2.5%)	Park and Sanders (1992)
Leafy vegetables	Malaysia	*Salmonella*	1/24 (4%)	Arumugaswamy *et al.* (1995)
	Malaysia	*L. monocytogenes*	5/22 (22.7%)	Arumugaswamy *et al.* (1994)
Leeks	Spain	*L. monocytogenes*	1/5 (20%)	de Simon *et al.* (1992)
Lettuce	Italy	*Salmonella*	82/120 (68%)	Ercolani (1976)
	Canada	*Campylobacter*	2/67 (3.1%)	Park and Sanders (1992)
	Canada	*L. monocytogenes*	3/15 (20%)	Odumeru *et al.* (1997)
	Lebanon	*Staphylococcus*	(14.3%)	Abdelnoor *et al.* (1983)
	Netherlands	*Salmonella*	2/28 (7.1%)	Tamminga *et al.* (1978)
	Saudi Arabia	*L. monocytogenes*		Salamah (1993)
	Saudi Arabia	*Y. enterocolitica*		Salamah (1993)
	Spain	*Salmonella*		Garcia-Villanova Ruiz*etal.*(1987b)
	United States	*Aeromonas*		Callister and Agger (1989)
Mungbean sprouts	United States	*Salmonella*		O'Mahony *et al.* (1990)
Mushrooms	United States	*C. jejuni*	3/200 (1.5%)	Doyle and Schoeni (1986)
Mustard cress	United Kingdom	*Salmonella*		Joce *et al.* (1990)
Mustard sprouts	United States	*B. cereus*		Portnoy *et al.* (1976)
	Canada	*Campylobacter*	1/42 (2.4%)	Park and Sanders (1992)
Parsley	Egypt	*Shigella*	1/250 (0.4%)	Satchell *et al.* (1990)
	Lebanon	*Staphylococcus*	(7.7%)	Abdelnoor *et al.* (1983)
	Spain	*Salmonella*	1/23 (4.3%)	Garcia-Villanova Ruiz*etal.*(1987b)
Pepper	Canada	*L. monocytogenes*	1/15 (6.7%)	Odumeru *et al.* (1997)
	Sweden	*Salmonella*		Andersson *et al.* (1989)
	United States	*C. botulinum*	1/201 (0.5%)	Lilly *et al.* (1996)
	United States	*Aeromonas*		Callister and Agger (1989)

Vegetale	Country	Pathogen	Prevalence	Peference
Potatoes	Saudi Arabia	*L. monocytogenes*		Salamah (1993)
	Saudi Arabia	*Y. enterocolitica*		Salamah (1993)
	Spain	*L. monocytogenes*	2/12 (16.7%)	de Simon *et al.* (1992)
	United States	*L. monocytogenes*	19/70 (27.1%)	Heisick *et al.* (1989a)
	United States	*L. monocytogenes*	28/132 (21.1%)	Heisick *et al.* (1989b)
	Canada	*Campylobacter*	1/63 (1.6%)	Park and Sanders (1992)
Prepacked salads	Northern Ireland	*L. monocytogenes*	3/21 (14.3%)	Harvey and Gilmour (1993)
	United Kingdom	*L. monocytogenes*	4/60 (13.3%)	Sizmur and Walker (1988)
	United Kingdom	*L. monocytogenes*		Velani and Roberts (1991)
Radish	Lebanon	*Staphylococcus*	(6.3%)	Abdelnoor *et al.* (1983)
	Saudi Arabia	*L. monocytogenes*		Salamah (1993)
	Saudi Arabia	*Y. enterocolitica*		Salamah (1993)
	United States	*L. monocytogenes*	25/68 (36.8%)	Heisick *et al.* (1989a)
	Canada	*Campylobacter*	2/74 (2.7%)	Park and Sanders (1992)
	United States	*L. monocytogenes*	19/132 (14.4%)	Heisick *et al.* (1989b)
Salad greens	Egypt	*Salmonella*	1/250 (0.4%)	Satchell *et al.* (1990)
	United Kingdom	*S. aureus*	13/256 (5.1%)	Houang *et al.* (1991)
Salad vegetables	Canada	*L. monocytogenes*	6/15 (40%)	Odumeru *et al.* (1997)
	Egypt	*Shigella*	3/250 (1.2%)	Satchell *et al.* (1990)
	Egypt	*Saureus*	3/36 (8.3%)	Satchell *et al.*(1990)
	Spain	*Aeromonas*	2/33 (6.1%)	Garcia-Gimeno *et al.* (1996a)
	Spain	*L. monocytogenes*	21/70 (30%)	Garcia-Gimeno *et al.* (1996b)
	United States	*Staphylococcus*		Harris *et al.* (1975)
	Germany	*L. monocytogenes*	6/263 (2.3%)	Breer and Baumgartner (1992)
	Northern Ireland	*L. monocytogenes*	4/16 (25%)	Harvey and Gilmour (1993)
	United States	*C. botulinum*	2/82 (2.4%)	Lilly *et al.* (1996)
	United Kingdom	*Y. enterocolitica*		Brockelhurst et al. (1987)
Seed sprouts	Canada	*Staphylococcus*	13/54 (24%)	Prokopowich and Blank (1991)
Soybean sprouts	United States	*B. cereus*		Portnoy *et al.* (1976)
Spinach	Canada	*Campylobacter*		Park and Sanders (1992)
	Spain	*Salmonella*	2/60 (3.3%)	Garcia-Villanova Ruiz*etal.*(1987b)
	United States	*Aeromonas*	2/38(5.2%)	Callister and Agger (1989)
Sprouting seeds	United States	*B. cereus*	56/98 (57%)	Harmon *et al.* (1987)
Tomato	Pakistan	*L. monocytogenes*	2/15 (13.3%)	Vahidy (1992)
Vegetables	Egypt	*Salmonella*	2/250 (0.8%)	Satchell *et al.* (1990)
	France	*Y. enterocolitica*	4/58 (7%)	Catteau *et al.* (1985)
	France	*Y. enterocolitica*	15/30 (50%)	Darbas *et al.*(1985)
	Iraq	*Salmonella*	3/43 (7%)	Al-Hindawi and Rished (1979)
	Italy	*L. monocytogenes*	7/102 (6.9%)	Gola *et al.* (1990)
	Italy	*Y. enterocolitica*	1/102 (1.0%)	Gola *et al.* (1990)
	Spain	*L. monocytogenes*	8/103 (7.8%)	de Simon *et al.* (1992)
	Spain	*Salmonella*	46/849 (5.4%)	Garcia-Villanova Ruiz*etal.*(1987a)
	Taiwan	*L. monocytogenes*	6/49 (12.2%)	Wong *et al.* (1990)
	United Kingdom	*L. monocytogenes*	4/64 (6.2%)	MacGowan *et al.* (1994)
	United States	*Salmonella*	4/50 (8.0%)	Rude *et al.* (1984)

[1] Adapted from Beuchat (1996b), with permission from the International Association of Milk, Food and Environmental Sanitarians. Most vegetables were grown in the country in which they were analysed for the presence of pathogens.

addition, inadequate water resources may encourage the use of unsafe water to make ice or raw sewage to irrigate fields. Farm environments are not and cannot be aseptic. Reduction in the chances of contamination can be achieved, however, through appropriate agronomic practices, harvesting, processing, shipping, marketing and preparation.

The use of properly composted manure and properly treated irrigation and spray waters, as well as pathogen-free water for washing, and for ice, will minimize the risk of contamination of fruits and vegetables with microbial pathogens. Good hygienic practice during production and transport, including sanitizing of harvesting equipment and transport vehicles, as well as the application of good hygienic practice during processing and preparation are critical. Elimination of animals and insects from processing, storage, marketing and food-service facilities should be a goal of anyone who handles raw produce. The highest level of hygiene must be practised by all handlers (including consumers) of fruits and vegetables, from the field to the table, if any degree of success is to be achieved in minimizing the risk of contamination.

The microorganisms normally present on the surface of raw fruits and vegetables may consist of chance contaminants from the soil or dust, or bacteria or fungi that have grown and colonized by utilizing nutrients exuded from plant tissues. Among the groups of bacteria commonly found on plant vegetation are those that test positive for coliforms or faecal coliforms — e.g. *Klebsiella* and *Enterobacter* (Duncan and Razzell, 1972; Splittstoesser *et al.*, 1980; Zhao *et al.*, 1997). Thus, the presence of coliforms or faecal coliforms on raw fruits and vegetables does not necessarily provide an index of faecal contamination. Microorganisms capable of causing human disease can, however, be found on raw produce (Table 1), and should be viewed as a threat to public health (Beuchat, 1996b; Doyle, 1990). What is not well known is where contamination occurs along the farm-to-fork pathway. Means to address this issue must address the entire system from production to consumption.

The presence of pathogenic and other microorganisms on fruits and vegetables is dictated not so much by surface topography as by exposure to environmental factors that lead to contamination. Although rough, highly-textured surfaces with deep crevices would be more likely to harbour soil, with the possible consequence of increased numbers of microorganisms, than would smooth-surfaced fruits and vegetables, differences in microbial profiles result largely from unrelated factors such as resident microflora in the soil, application of non-resident microflora via animal manures, sewage or irrigation water, rainfall and atmospheric humidity. The presence of a pathogen on produce is of less consequence if the rind, skin or peel is to be removed before consumption. Bananas, melons, mangoes, durians, pineapples and papayas, for example, fall in this category. However, the process of removing the rind, skin or peel from these fruits, perhaps with the exception of bananas, may result in contamination of the edible portion, thereby creating a risk to the consumer. Microorganisms that have become trapped on the inner leaves of certain vegetables can be particularly difficult to remove by routine cleansing practices. Brief descriptions of pathogens that have been isolated from raw fruits and vegetables are given below, with special emphasis on their association with outbreaks of human disease.

Table 2. Examples of pathogens associated with fruits and vegetables involved in outbreaks of foodborne disease

Agent	Implicated/ suspected food	Reference
Bacillus cereus	sprouts	Portnoy *et al.* (1976)
Campylobacter	cucumber	Kirk *et al.* (1997)
Campylobacter jejuni	lettuce	CDC (1998)
Clostridium botulinum	vegetable salad	PHLS (1978)
Cryptosporidium	apple cider	CDR (1991)
Cyclospora	raspberries	Herwaldt *et al.*(1997)
E.coli O157	radish sprouts	WHO (1996)
E.coli O157	apple juice	CDC (1996)
E.coli O157	apple cider	Besser *et al.* (1993)
E.coli O157	iceberg lettuce	CDR(1997)
Fasciolia hepatica	watercress	Hardman (1970)
Giardia	vegetables, incl.carrots	Mints *et al.* (1992)
Hepatitis A virus	iceberg lettuce	Rosenblum *et al.*(1990)
Hepatitis A virus	raspberries	Ramsay *et al.* (1989)
Hepatitis A virus	strawberries	Niu *et al.* (1992)
Norwalk virus	tossed salad	Lieb *et al.* (1985)
Salmonella Agona	coleslaw & onions	Clark *et al.* (1973)
Salmonella Miami	watermelon	Gayler *et al.* (1955)
Salmonella Oranienburg	watermelon	CDC (1979)
Salmonella Poona	cantaloupes	CDC (1991)
Salmonella Saint-Paul	beansprouts	O'Mahony *et al.* (1990)
Salmonella Stanley	alfalfa sprouts	Mahon *et al.*(1997)
Salmonella Thompson	root vegetables & dried seaweed	Kano *et al.*(1996)
Shigella flexneri	mixed salad	Dunn *et al.* (1995)
Shigella sonnei	iceberg lettuce	Kapperud *et al.* (1995)
Shigella sonnei	tossed salad	Martin *et al.* (1986)
Vibrio cholerae	salad crops & vegetables	Shuval, *et al.* (1989)

Pathogens of most concern

Salmonella

The antigenic scheme for classifying salmonellae recognizes more than 2300 serovars and, while all can be considered human pathogens, only about 200 are associated with human illness. Animal husbandry practices used in the poultry, meat and fish industries, and the recycling of offal and inedible raw materials into animal feeds, has favoured the continued prominence of *Salmonella* in the global food chain (D'Aoust, 1997). There are reports of human salmonellosis linked to cantaloupe (Ries *et al.*, 1990) and sprouts produced from alfalfa seeds (Mahon *et al.*, 1996) imported to the United States. Hygienic conditions during the production, harvesting, transport and distribution of raw fruits and vegetables from some countries may not always meet minimum hygienic requirements, thus facilitating contamination on arrival in another country. Application of night soil, untreated sewage sludge or effluents, or irrigation water containing untreated sewage to fields and gardens can result in contamination of fruits and vegetables with *Salmonella* and other pathogens. Washing fruits and vegetables with contaminated water and handling of produce by infected workers, vendors and consumers in the marketplace helps the spread of pathogenic microorganisms, including *Salmonella*.

Salmonellae have been isolated from many types of raw fruits and vegetables (Beuchat, 1996b; Wells and Butterfield, 1997). Outbreaks of salmonellosis have been linked to a diversity of fruits and vegetables, including tomatoes (Centers for Disease Control and Prevention, 1993; Hedberg *et al.*, 1993; Wood *et al.*, 1991), bean sprouts (Mahon *et al.*, 1996; O'Mahony *et al.*, 1990; Van Venedey *et al.*, 1996), melons (Blostein, 1991; Centers for Disease Control and Prevention, 1979; 1991; Gaylor *et al.*, 1955; Ries *et al.*, 1990), unpasteurized orange juice (Cook *et al.*, 1990) and apple juice (Centers for Disease Control and Prevention, 1975). The pathogen can grow on the surface of alfalfa sprouts (Jaquette *et al.*, 1996), tomatoes (Zhuang *et al.*, 1995) and perhaps on other mature raw fruits and vegetables, making it imperative to use hygienic practices when handling them.

Shigella

Bacillary dysentery or shigellosis is caused by *Shigella*, of which there are four species: *S. dysenteriae, S. flexneri, S. boydii* and *S. sonnei* (Maurelli and Lampel, 1997). Most cases of shigellosis result from the ingestion of food or water contaminated with human faeces. Like salmonellae and other pathogens present in faeces, *Shigella* can contaminate raw fruits and vegetables by several routes, including insects and the hands of persons who handle the produce, although shigellosis is more often transmitted from person to person.

Several large outbreaks of shigellosis have been attributed to the consumption of contaminated raw fruits and vegetables. Lettuce (Davis *et al.*, 1988; Frost *et al.*, 1995; Kapperud *et al.*, 1995; Martin *et al.*, 1986), scallions (Cook *et al.*, 1995), vegetable salad (Dunn *et al.*, 1995), potato salad containing spring onions (Formal *et al.*, 1965), salad vegetables (Public Health Research Service, 1997) and watermelon (Frelund *et al.*, 1987) have been implicated as vehicles of shigellosis. Sliced raw papaya, jicama and watermelon support the growth of *Shigella* species (Escartin *et al.*, 1989). *S. sonnei* survived on refrigerated, shredded lettuce for three days without decreasing in number, and increased when held at 22°C (Satchell *et al.,* 1990).

Escherichia coli

Escherichia coli is common in the normal microflora of the intestinal tracts of humans and other warm-blooded animals. Strains that cause diarrhoeal illness are categorized into groups on the basis of virulence properties, mechanisms of pathogenicity, clinical syndromes and antigenic characteristics. The major groups are designated as enterotoxigenic, enterohaemorrhagic, enteropathogenic, enteroinvasive, diffuse-adhering and enteroaggregative (Doyle *et al.*, 1997). Fruits and vegetables can become contaminated with one or more of these groups while in the field or during post-harvest handling. Sources and mechanisms of contamination are similar to those described for *Salmonella* and *Shigella.*

Enterotoxigenic *E. coli* is a cause of traveller's diarrhoea, an illness sometimes experienced when individuals visit countries with food and water hygiene standards different from their own. Contaminated raw vegetables are thought to be a common cause of traveller's diarrhoea. Illness has been associated with consumption of salads (Merson *et al.*, 1976; Mintz, 1994) and carrots (Centers for Disease Control and Prevention, 1994). Enterohaemorrhagic *E. coli* O157:H7 has more recently been recognized as a foodborne pathogen. Since cattle appear to be a natural reservoir for the pathogen, most outbreaks of illness have been associated with the consumption of contaminated, undercooked beef and dairy products. However, outbreaks have also been linked to lettuce (Ackers *et al.*, 1996; Mermin *et al.*, 1996), apple cider (Besser *et al.*, 1993; Centers for Disease Control and Prevention, 1996; Steele *et al.*, 1982), radish sprouts (Nathan, 1997) and alfalfa sprouts (Centers for Disease Control and Prevention, 1997e). Enterohemorrhagic *E. coli* can grow on cantaloupe and watermelon cubes (del Rosario and Beuchat, 1995), shredded lettuce (Diaz and Hotchkiss, 1996) and sliced cucumbers (Abdul-Raouf *et al.*, 1993), and in apple cider (Zhao *et al.*, 1993).

Contamination of raw fruits and vegetables with enterohaemorrhagic *E. coli* O157:H7 may occur when cattle, and perhaps other ruminants such as deer, inadvertently enter fields, or when improperly composted cow manure has been applied as a fertilizer. The potential for contamination may be enhanced when fruits or vegetables have fallen from the plant to the ground and are then picked and placed into the handling and processing chain. Also, because contaminated manure may become airborne dust particles, it is possible that fruits on trees and vines may become contaminated. Workers on farms and in packing houses may also be a source of *E. coli* O157:H7. These mechanisms of contamination are somewhat speculative at present and must be thoroughly investigated before appropriate interventions can be introduced to reduce the risk.

Campylobacter

Campylobacter jejuni is a leading cause of bacterial enteritis in many countries. Reservoirs of this pathogen include several wild animals as well as poultry, cows, pigs and domestic pets (Nachamkin, 1997). While consumption of food of animal origin, particularly poultry, is largely responsible for infection, *Campylobacter* enteritis has also been associated with the consumption of raw fruits and vegetables (Bean and Griffin, 1990; Harris *et al.*, 1996). Although *Campylobacter* does not grow at temperatures below 30 °C and is sensitive to acid pH, it can survive on cut fruits for sufficient time to be a risk to the consumer (Castillo and Escartin, 1994).

Yersinia enterocolitica

Yersinia enterocolitica can be found in a variety of terrestrial and freshwater ecosystems, including soil, vegetation and water in lakes, rivers, wells and streams (Kapperud, 1991), but most isolates from these sources lack virulence for humans. Pigs, however, frequently carry serotypes capable of causing human disease. The ability of *Y. enterocolitica* to grow at refrigeration temperature and its documented presence on raw produce raises concern about the potential of salad vegetables as causative vehicles of yersiniosis in humans. Seven per cent of carrot samples obtained from eating establishments in France were reported to contain serotypes of *Yersinia* that may be pathogenic to humans (Catteau *et al.*, 1985). In another study (Darbas *et al.*, 1985), 50% of raw vegetables analysed contained nonpathogenic strains of *Yersinia*. Incidence was higher on root and leafy vegetables than on tomatoes or cucumbers. Certainly, application of improperly composted pig manure to vegetable fields should be avoided to reduce the possibility of pathogenic strains being present on produce when it reaches the consumer.

Listeria monocytogenes

Listeria monocytogenes is present in the intestinal tract of many animals, including humans, so it is not surprising that the organism can also be found in the faeces of these animals, on the land they occupy, in sewage, in soils to which raw sewage is applied and on plants which grow in these soils (Van Renterghem *et al.*, 1991). The organism also exists in nature as a saprophyte, growing on decaying plant materials, so its presence on raw fruits and vegetables is not rare (Beuchat, 1992; 1996a; Beuchat *et al*, 1990). Surveys of fresh produce have revealed its presence on cabbage, cucumbers, potatoes and radishes in the United States (Heisick *et al.*, 1989), ready-to-eat salads in the United Kingdom (Sizmur and Walker, 1988), the Netherlands (Beckers *et al.*, 1989), Northern Ireland (Harvey and Gilmour, 1993) and Canada (Odumeru *et al.*, 1997), tomatoes and cucumbers in Pakistan (Vahidy, 1992), and bean sprouts, sliced cucumbers and leafy vegetables in Malaysia (Arumugaswamy *et al.*, 1994).

A cabbage-associated outbreak of listeriosis has been documented (Schlech *et* al., 1983). *Listeria monocytogenes* grows at temperatures as low as 2°C (Rocourt and Cossart, 1997) and can thrive in cool, wet areas in processing facilities. It can also grow on endive (Carlin, 1994; Carlin *et al.*, 1995), lettuce (Beuchat and Brackett, 1990a), tomatoes (Beuchat and Brackett, 1991), asparagus, broccoli and cauliflower (Berrang *et al.*, 1989b) and cabbage (Beuchat *et al.*, 1986) but appears to be inhibited by carrot juice (Beuchat and Brackett, 1990b; Beuchat *et al.*, 1994; Beuchat and Doyle, 1995; Nguyen-the and Lund, 1991; 1992). Production of 6-methoxymellein and 6-hydroxymellein by carrot cells infected by fungi or upon partial hydrolysis is known to occur (Amin *et al.*, 1986; Kurusaki and Nishi, 1984). These phytoalexins inhibit a wide range of spoilage and pathogenic bacteria. The mechanism of action of 6-methoxymellein apparently involves interference with membrane-associated functions. Controlled atmosphere storage has been shown to extend the shelf-life of broccoli and asparagus but does not influence the rate of growth of *L. monocytogenes* (Berrang *et al.*, 1989b). The risk of listeriosis increases when these vegetables are stored for longer periods before consumption because *L. monocytogenes* has a greater opportunity to increase.

Staphylococcus aureus

Staphylococcus aureus is known to be carried in the nasal passages of healthy food handlers and has been detected on raw produce (Abdelnoor *et al.*, 1983) and ready-to-eat vegetable salads

(Houang *et al.*, 1991). However, enterotoxigenic *S. aureus* does not compete well with other microorganisms normally present on raw fruits and vegetables, so spoilage caused by nonpathogenic microflora would probably precede the development of the high populations of this pathogen that would be needed for production of staphylococcal enterotoxin.

Clostridium species

Spores of *Clostridium botulinum* and *Clostridium perfringens* can be found both in soil and on raw fruits and vegetables. The high rate of respiration of salad vegetables can create an anaerobic environment in film-wrapped packages, thus favouring the growth of *C. botulinum* and botulinal toxin production. Botulism has been linked to coleslaw prepared from packaged, shredded cabbage (Solomon *et al.*, 1990) and chopped garlic in oil (St. Louis *et al.*, 1988). Studies have revealed that *C. botulinum* can produce toxin in polyvinyl film-packaged (Sugiyama and Yang, 1975) and vacuum-packaged mushrooms (Malizio and Johnson, 1991). It is important that the permeability characteristics of packaging films minimize the possibility of development of anaerobic conditions suitable for outgrowth of clostridial spores. Recognizing that anaerobic pockets may develop in tightly packed produce, even when films have high rates of oxygen and carbon dioxide permeability, an additional measure to prevent growth of *C. botulinum* is to store produce at less than 3°C.

Bacillus cereus

Spores of enterotoxigenic strains of *Bacillus cereus* are common in most types of soil. Some strains can grow at refrigeration temperatures. Foods other than raw fruits and vegetables are generally linked to illness implicating *B. cereus*. Illness associated with eating contaminated soy, mustard and cress sprouts has, however, been documented (Portnoy *et al.*, 1976). Human illness tends to be restricted to self-limiting diarrhoea (enterotoxin) or vomiting (emetic toxin). However, emetic toxin-producing strains have produced liver failure and death by the foodborne route.

Vibrio species

Vibrio species are generally the predominant bacterial species in estuarine waters and are therefore associated with a great variety of fish and seafoods. There are 12 human pathogenic *Vibrio* species, of which *Vibrio cholerae, V. parahaemolyticus* and *V. vulnificus* are of greatest concern (Oliver and Kaper, 1997). *Vibrio cholerae* is the causative agent of cholera, one of the few foodborne diseases with epidemic and pandemic potential. Carriage of the organism by infected humans is important in transmission of disease. Water can become contaminated by raw sewage. Ingestion of water containing *V. cholerae* or of foods that are washed with contaminated water but not disinfected can lead to widespread transmission of cholera (Mintz *et al.*, 1994). An outbreak of cholera linked to the consumption of unpasteurized coconut milk has been documented (Taylor *et al.*, 1993).

Vibrio parahaemolyticus is perhaps the best described of the pathogenic vibrios in terms of its involvement in foodborne illness. Outbreaks are generally associated with consumption of contaminated raw or undercooked seafoods and are seasonal, peaking in summer when *V. parahaemolyticus* is at its highest population in estuarine water. Cross-contamination of raw fruits and vegetables with seafoods during handling, particular at retail locations, represents a potential mode of transmission to humans.

Of all the vibrios, *V. vulnificus* infection from contaminated seafoods results in the highest fatality rate, typically 60% in individuals suffering from liver diseases (Oliver and Kaper, 1997). As with *V. cholerae* and *V. parahaemolyticus*, it is possible that cross-contamination of raw fruits and vegetables with *V. vulnificus* could occur, resulting in human infection when they are consumed.

Viruses

Viruses can be excreted in large numbers by infected individuals (Cliver, 1997). Although viruses will not grow in or on foods, raw fruits and vegetables may serve as vehicles for infection. Many food-associated outbreaks of hepatitis A have been recorded (Cliver, 1997). In most instances, these outbreaks have not appeared to depend on the stability of the virus in the food. Shellfish taken from waters contaminated with human faeces have been the vehicle in most outbreaks, but any food handled by an infected person may become contaminated and transmit infection (Cliver, 1985). Hepatitis A infection has been linked to the consumption of lettuce (Rosenblum *et al.*, 1990), diced tomatoes (Williams *et al.*, 1994), raspberries (Ramsay and Upton, 1989; Reid and Robinson, 1987) and strawberries (Centers for Disease Control and Prevention, 1997a; Niu *et al.*, 1992). Hernandez *et al.* (1997) suggested that lettuce contaminated with sewage could be a vehicle for hepatitis A virus and rotavirus. Lettuce obtained from farmers' markets was reported to contain hepatitis A virus. The extent to which hepatitis A and other viruses are removed from the surface of fruits and vegetables upon treatment with chemical disinfectants is not known.

The number of cases of foodborne disease caused by Norwalk-like viruses (i.e. Small Round Structured Viruses, or SRSV) appears to be on the increase (Bean and Griffin, 1990). Outbreaks have a pattern of transmission resembling that of hepatitis A. Ice made from contaminated water has been implicated as the vehicle in more than one outbreak but salad items have also been linked to Norwalk-like gastroenteritis (Karitsky *et al.*, 1995). Workers who have prepared salads linked to viral gastroenteritis have been shown to have high antibody titers to Norwalk virus (Griffin *et al.*, 1982; Gross *et al.*, 1989; Iverson *et al* 1987). A non-typical outbreak of Norwalk virus gastroenteritis associated with exposure of celery to nonpotable water has been reported (Warner, 1991). Studies have shown that viruses may persist for weeks or even months on vegetable crops and in soils that have been irrigated or fertilized with sewage wastes (Larkin *et al.*, 1978).

Rotaviruses, astroviruses, enteroviruses (polioviruses, echoviruses and coxsackie viruses), parvoviruses, adenoviruses and coronaviruses have been reported to be transmitted by foods on occasion (Cliver, 1994). At least one echovirus outbreak has been attributed to contaminated raw shredded cabbage (New York Department of Health, 1989).

Parasites

The major modes of transmission of protozoa include consumption of surface water, exposure to contaminated recreational water, animal-to-person contact and person-to-person contact (Speer, 1997). However, the epidemiology of protozoa most commonly associated with human infections, namely *Giardia, Entamoeba, Toxoplasma, Sarcocystis*, *Isopora*, *Cryptosporidium, Eimeria* and *Cyclospora*, is not fully understood (Goodgame, 1996). While the life cycles of each of these parasites differ, all require passage through an animal or human host. Shedding of cysts or spores into faeces which may then, directly or indirectly (e.g. via sewage or irrigation water), contaminate raw fruits and vegetables occurs on a global scale; it may be common in countries where hygienic conditions (especially water quality) are compromised. Expanding poverty, urban migration, food exportation, air travel and immigrant populations in conjunction with other societal and environmental changes make the distinction between so-called tropical and western parasitic diseases less clear-cut than in the past.

Outbreaks of protozoan infections in humans have been linked to raw fruits and vegetables. Epidemiological evidence has implicated an asymptomatic food handler as the probable source of *Giardia lamblia* and raw sliced vegetables as the vehicle of transmission in an outbreak of giardiasis (Mintz *et al,* 1993). Raspberries (Centers for Disease Control and Prevention, 1996c; 1997b,c; Jackson *et al.*, 1997), lettuce (Centers for Disease Control and Prevention, 1997c), and basil (Centers for Disease Control and Prevention, 1997d) have been the implicated vehicles of transmission in outbreaks of *Cyclospora cayetanensis* infection, and unpasteurized apple juice has been linked to outbreaks of cryptosporidiosis caused by *Cryptosporidium parvum* (Millard *et al.*, 1994). A survey of vegetables has revealed the presence of *Cryptosporidium* oocysts on cilantro, lettuce, radish, tomato, cucumber and carrot (Monge and Chinchilla, 1996). The current status of knowledge about *Cryptosporidium* and its significance in food and beverage production has been reviewed by Donnelly and Stentiford (1997). The presence of these protozoa on raw fruits and vegetables is likely to be due to contact with animal or human faeces, sewage, water containing untreated sewage and sludge from primary or secondary municipal water treatment facilities. While *Cryptosporidium, Giardia* and other parasites in water are quite resistant to chlorine and other disinfectants, little is known about the efficacy of these disinfectants in killing or removing parasites from the surface or tissues of fruits and vegetables. Surveys have shown that there is a high incidence of the parasitic roundworm, *Ascraris*, in the sewage sludge of many cities (Jackson *et al.*, 1977).

Foodborne trematode infections are major health problems, with an estimated 40 million persons, mainly in eastern and southern Asia, being affected (Abdussalam *et al.*, 1995). Infection takes place through the consumption of raw plants, or raw or undercooked freshwater fish or shellfish, containing the infective cyst (metacercaria) stage of these parasites. Watercress is a major source of *Fasciola hepatica* infection (World Health Organization, 1995). Over 300,000 clinical cases of fascioliasis may have occurred in more than 55 countries in Africa, the Americas, Asia, Europe and the western Pacific from 1970 to 1990 (Chen and Mott, 1990). Large endemic areas have been reported more recently in Bolivia, Egypt, Iran and Peru (Abdussalam *et al.*, 1995).

A wide range of other aquatic plants may support metacercariae. Conditions for transmission of *Fasciolopsis buski* are present in areas of cultivation of water caltrop, water chestnut, water hyacinth, water bamboo, water mimosa, lotus and duckweed. If animal manure or effluent from livestock pens or abattoirs is used as fertilizer for these plants, it introduces *Fasciolopsis* to the aquatic environment. Plantborne trematodes encyst as metacerceriae on the surface of plants or on debris floating on the water surface. Plants that grow in water are also believed to serve as hosts for encystment of metacercariae of certain intestinal flukes (World Health Organization, 1995). Non-aquatic plants such as lettuce, alfalfa, mint and sugarcane which may be eaten raw have also been implicated in human trematode infections.

Strategies for controlling foodborne trematode infections associated with eating raw plant materials will be shaped by ecological and environmental factors. The use of non-composted animal manures or effluent from livestock pens for the purpose of fertilizing aquatic plants should cease. Freezing, heating and irradiation can be effective in killing trematodes. The effectiveness of chemical treatments in killing trematodes attached to plant surfaces is not well understood.

Mitigating the risk of disease

To minimize the risk of infection or intoxication associated with raw fruits and vegetables, potential sources of contamination from the environment to the table should be identified and specific measures and interventions to prevent and/or minimize the risk of contamination should be considered and correctly implemented. Where the possibility of contamination cannot be excluded, the application of the most effective decontamination processes should be considered. In many regions of the world, present practices in agriculture practice cannot assure fruits and vegetables that are free from pathogens, as the studies mentioned in Table 1 illustrate. Application of good hygienic practice during production, transport and processing, combined with the Hazard Analysis Critical Control Point system, will certainly minimize the contamination of fruits and vegetables and reduce the risk of illness associated with these foods. However, food handlers and consumers also need to observe good hygienic practice during processing and preparation of these foods for consumption — including treatments for reducing the number of pathogens.

The simple practice of washing raw fruits and vegetables in hot water or water containing detergent or permanganate salts removes a portion of the pathogenic and spoilage microorganisms that may be present, but studies showing the efficacy of these treatments are few. Even washing fruits and vegetables in potable water, then again washing or rinsing in potable water would aide in removing microorganisms. Additional 10-fold to 100-fold reductions can sometimes be achieved by treatment with disinfectants. Viruses and protozoan cysts on fruits and vegetables generally exhibit higher resistance to disinfectants than do bacteria or fungi. However, relative resistance varies greatly with the type and pH of disinfectant, contact time, temperature and the chemical and physical properties of the fruit or vegetable surface. Little is known about the efficacy of disinfectants in relation to the roughness of fruit and vegetable surfaces, although higher amounts of cuticle material may protect against embedding of cells, thereby increasing the need for exposure to chemical treatments.

Several types of treatment are known to be partially effective in removing disease-causing organisms from the surface of whole and cut raw fruits and vegetables or from contact surfaces during handling. Perhaps with the exception of irradiation, none of these treatments can be relied upon to totally disinfect raw produce, at least when administered at levels that will not cause deterioration in sensory quality. Even irradiation may not be completely effective in killing viruses on fruits and vegetables. Rather, these treatments should be considered as methods of disinfection, causing reductions in populations of microorganisms but not always yielding fruits and vegetables free of pathogens.

Each type of dis infectant has its own efficacy in killing microbial cells. Effectiveness depends on the nature of the cells as well as the characteristics of fruit and vegetable tissues and juices. Some types of disinfectants are appropriate for use in direct contact washes, while others are suitable only for equipment or containers used to process, store or transport fruits and vegetables. The mechanism of action of many disinfectants on microbial cells and the influence of factors associated with plant materials is poorly understood.

The legal use of various treatments differs from country to country. Described here are disinfectants and disinfection treatments that are in current use or that have potential for killing microorganisms on whole and cut raw fruit and vegetables, as well as surfaces that are in contact with produce during handling.

Chlorine

Chlorine has been used for many years to treat drinking-water and wastewater as well as to sanitize food processing equipment and surfaces in processing environments. It is also used as a disinfectant in wash, spray and flume waters in the raw fruit and vegetable industry. Chlorination is generally accomplished by using elemental chlorine or one of the hypochlorites. Liquid chlorine and hypochlorites are moderately effective disinfectants for surfaces that may come in contact with fruits and vegetables during harvesting and handling, for processing equipment, and for whole and cut fruits and vegetables. To disinfect produce, chlorine is commonly used at concentrations of 50-200 ppm with a contact time of 1-2 minutes.

Inhibitory or lethal activity depends on the amount of free available chlorine (as hypochlorous acid, or HOCl) in the water that comes in contact with microbial cells. The dissociation of HOCl depends on the pH, and chlorine is consumed on contact with organic matter. As the pH of the solution is reduced, the equilibrium is in favour of HOCl. However, since metal containers and processing equipment are often susceptible to corrosion at low pH, a pH of 6.0-7.5 is most appropriate for effective sanitizing activity without damaging equipment surfaces. The percentages of chlorine as HOCl at pH 6.0 and 8.0 are about 97% and 23%, respectively, at 20°C. Toxic chlorine gas (Cl_2) is formed at a pH below 4. At a given pH, equilibrium is in favour of HOCl as the temperature is decreased. This is because chlorine vaporizes as water temperature increases. Chlorine rapidly loses activity on contact with organic matter or exposure to air, light or metals. A concern among people who use chlorinated water as a disinfectant is that prolonged exposure to chlorine vapours can cause irritation to the skin and respiratory tract. The occupational exposure limit (ceiling) is 1 ppm in the United States (instantaneous up to 15 minutes) (OSHA). The formation of potentially hazardous organochlorines upon treatment of fruits and vegetables with chlorine is also a possible concern, although these have not been characterized.

Maximum solubility of chlorine is achieved in water at about 4°C. However, the temperature of the chlorinated water should ideally be at least 10°C higher than that of fruits or vegetables to achieve a positive temperature differential, thereby minimizing the uptake of wash-water through stem tissues (Bartz and Showalter, 1981; Zhuang *et al.*, 1995) and open areas in the skin or leaves, whether due to mechanical assault or naturally present (e.g. lenticels and stomata). Elimination of uptake of wash-water that may contain microorganisms, including those that may cause human illnesses, should be considered as a critical control point in handling, processing and disinfection of raw fruits and vegetables.

Possible uses of chlorinated water in packing-houses and during washing, cooling and transport for the purpose of controlling post-harvest diseases of fruits and vegetables have been reviewed by Eckert and Ogawa (1988). The effects of chlorine concentration on aerobic microorganisms and faecal coliforms present on leafy salad greens was studied by Mazollier (1988). Populations of pathogens were markedly reduced with increased concentrations of chlorine to 50 ppm, but further increases in concentration to 200 ppm did not have a substantial additional effect. A standard procedure for washing lettuce leaves in tap-water reduced populations (ca. 10^7/g) of microflora by 92% (Adams *et al.*, 1989). Inclusion of 100 ppm chlorine (pH 9.0) reduced the count by 97.8%, indicating that this concentration of chlorine in treatment water was only slightly more effective than using water with no chlorine. Adjusting the pH from 9 to 4.5-5.0 with inorganic and organic acids resulted in a 1.5-4.0-fold increase in microbicidal effect. Increasing the washing time in hypochlorite solution from 5 to 30 minutes did not decrease numbers of microbes further, whereas extended washing in tap-water resulted in a reduction comparable to hypochlorite. The addition of 100 ppm of a surfactant (Tween 80) to hypochlorite washing solution enhanced lethality by enhancing surface contact but adversely affected sensory qualities of lettuce. Somers (1963) reported that wash-water with about 5 ppm chlorine reduced microbial populations on several fruits and vegetables by less than 90% from initial populations of 10^4-10^6 CFU/g.

In addition to the influence of pH and temperature on the effectiveness of chlorine in killing microorganisms that occur naturally on fruits and vegetables, the type of produce and diversity of microorganisms can also greatly influence efficacy. Garg *et al.* (1990), for example, observed that dipping lettuce in water containing 300 ppm chlorine reduced total microbial counts by about 1000-fold on lettuce but had no effect on microbial counts on carrots or red cabbage. Treatment of whole and shredded lettuce leaves in water containing 200-250 ppm chlorine reduced populations of aerobic microorganisms by 90-99% and psychotrophic microorganisms and yeasts and moulds by 50-90%.

Studies have aimed to determine the effectiveness of chlorine in killing bacterial pathogens inoculated onto the surface of raw vegetables. Dipping Brussels sprouts into a 200-ppm chlorine solution for 10 seconds decreased the number of viable *Listeria monocytogenes* cells (10^6 CFU/g) by about 100-fold (Brackett, 1987). However, dipping inoculated sprouts in sterile water containing no chlorine reduced the number of viable cells by about 10-fold. The maximum $\log_{10}$ reduction of *L. monocytogenes* on shredded lettuce and cabbage treated with 200 ppm chlorine for 10 minutes was reported to be 1.3-1.7 $\log_{10}$ CFU/g and 0.9-1.2 $\log_{10}$ CFU/g respectively (Zhang and Farber, 1996). Initial populations ranged from $\log_{10}$ 5.4 to 5.7 CFU/g. Reductions were greater when treatment was at 22°C than at 4°C, perhaps because the temperature of lettuce and cabbage was less than 22°C but more than 4°C. As noted above, the effectiveness of chlorine in killing microorganisms is generally greater if the temperature of the treatment solution is higher than the temperature of the fruit or vegetable. Treatment was more effective at both temperatures against *L. monocytogenes* on lettuce than on cabbage, indicating that antimicrobial activity is influenced by the nature of the vegetable being treated and/or the microflora it harbours. Numbers decreased only marginally with increased exposure time from 1 to 10 minutes, which agrees with observations by Brackett (1987) that the action of chlorine against *L. monocytogenes* occurs primarily during the first 30 seconds of exposure. Nguyen-the and Carlin (1994) concluded that the reduction of *L. monocytogenes* from the surface of vegetables by chlorine is both unpredictable and limited.

The efficacy of chlorine treatment on inactivation of *Salmonella* Montevideo on mature green tomatoes has been studied. Populations on the surface and in the stem core tissue were significantly reduced by dipping tomatoes for 2 minutes in a solution containing 60 ppm or 110 ppm chlorine respectively. However, treatment in a solution containing 320 ppm chlorine did not result in complete inactivation (Zhuang *et al.*, 1995). The ineffectiveness of 100 ppm chlorine against *S.* Montevideo inoculated into cracks in the skin of mature green tomatoes was demonstrated by Wei *et al.* (1995).

Immersion of warm (26-40°C) tomatoes for 10 minutes or longer in cool (20-22°C) suspensions of bacteria resulted in infiltration of cells into the stem tissue (Bartz and Showalter, 1981). Uptake of bacterial cells was associated with a negative temperature difference between the water and the tomato, i.e. the water temperature was less than the tomato temperature. When the differential was shifted to a positive relationship, i.e. when the water temperature was higher than the tomato temperature, the extent of infiltration was reduced. A significantly higher number of *S.* Montevideo cells has been shown to be taken up by the core tissue when tomatoes at 25°C are dipped in suspensions at 10°C compared with the number of cells taken up by tomatoes dipped in suspensions at 25 or 37°C (Zhuang *et al.*, 1995). Thus, the concentration of free chlorine reaching viable cells of *S.* Montevideo that have infiltrated the core tissues is reduced to the point that lethality is substantially diminished. The uptake of pathogens and spoilage microorganisms by other fruits and vegetables during washing has not been investigated. However, infiltration of microbial cells due to a negative temperature differential between the water and the fruit or vegetable would appear to be possible.

The effectiveness of chlorine in killing salmonellae on alfalfa seeds has been studied. Treatment of seeds inoculated with *Salmonella* Stanley (10^2-10^3 CFU/g) in 100 ppm chlorine solution for 10 minutes has been reported to cause a significant reduction of the pathogen; treatment in 290 ppm chlorine solution resulted in a significant reduction compared to treatment with 100 ppm chlorine (Jaquette *et al.*, 1996). However, solutions containing free chlorine concentrations up to 1000 ppm failed to result in further significant reductions. Treatment of seeds containing 10^1-10^2 CFU of *S.* Stanley per g for 5 minutes in a solution containing 2040 ppm chlorine reduced the population to <1 CFU/g. The sensory quality of sprouts produced from seeds receiving this treatment is not adversely affected.

In another study, alfalfa sprouts inoculated with a five-serovar (*S.* Agona, *S.* Enteritidis, *S.* Hartford, *S.* Poona, *S.* Montevideo) mixture of *Salmonella* were dipped in 200, 500 or 2000 ppm chlorine solutions for 2 minutes (Beuchat, 1997). The pathogen was reduced by about 3.4 $\log_{10}$ CFU/g after treatment with 500 ppm chlorine and to an undetectable level (<1 CFU/g) after treatment with 2000 ppm chlorine. Chlorine treatment (2000 ppm) of cantaloupe cubes inoculated with the same *Salmonella* serovars resulted in less than a 90% reduction in viable cells. The very high level of organic matter in the juice released from cut cantaloupe tissue apparently neutralizes the chlorine before its lethality can be manifested.

Failure to maintain adequate chlorine in wash-water may lead to increases in microbial populations on fruits and vegetables. In a study designed to determine microbiological changes in fresh market tomatoes during packing operations, Senter *et al.* (1985) observed that total plate counts and populations of Enterobacteriaceae were higher, compared to controls, on tomatoes washed in water containing an average of 114 ppm (range 90-140) chlorine; decreases were noted when tomatoes were treated in water containing 226 ppm chlorine (range 120-280). Recontamination of tomatoes occurred in the waxing operation as evidenced by increased total plate counts and mould populations.

Fruit and vegetable tissue components neutralize chlorine, rendering it inactive against microorganisms. The inaccessibility of hypochlorous acid to microbial cells in cracks, creases, crevices, pockets and natural openings in the skin undoubtedly also contributes to chlorine's overall lack of effectiveness. The hydrophobic nature of the waxy cuticle on the surface of fruits and vegetables protects microbial cells from exposure to chlorine and, undoubtedly, other chemicals used as disinfectants that do not penetrate or dissolve these waxes. Surface-active agents such as detergents and ethanol lessen the hydrophobicity of fruit and vegetable skins as well as the surfaces of edible leaves, stems and flowers, but also tend to cause deterioration of sensory qualities (Adams *et al.*, 1989; Zhang and Farber, 1996). Disinfectants that contain a solvent that would remove the waxy cuticle layer, and with it surface contaminants, without adversely affecting sensory characteristics would hold greater potential in reducing microbial populations on the surface of raw fruits and vegetables. Such disinfectants may be limited to use on fruits and vegetables that are to be further processed into juice or cut products, or on whole fruits, vegetables or plant parts that are destined for immediate consumption, since removal of cuticle material will also hasten deterioration of sensory quality. Application of such disinfectants at point of use diminishes the importance of this drawback.

Chlorine dioxide

Chlorine dioxide (ClO_2) has received attention as a disinfectant for fruits and vegetables, largely because its efficacy is less affected by pH and organic matter and it does not react with ammonia to form chloramines, as do liquid chlorine and hypochlorites. One disadvantage of ClO_2 is that it is unstable, it must be generated on site and can be explosive when concentrated. Chlorine dioxide decomposes at temperatures greater than 30°C when exposed to light. Compared to chlorine, fewer organohalogens are formed as reaction products of ClO_2. Its potential for use as a disinfectant has become more attractive in recent years due to the development of technologies that permit shipment to areas of use instead of requiring on-site generation. The oxidizing power of ClO_2 is reported to be about 2.5 times that of chlorine (Benarde *et al.*, 1967) and its activity is not affected by pH. Its mechanism of action involves disruption of cell protein synthesis and membrane permeability control.

In the United States, a maximum of 200 ppm ClO_2 is permitted for sanitizing equipment for fruit and vegetable processing. Chlorine dioxide is authorized for use in washing whole fresh fruits and vegetables and shelled beans and peas with intact cuticles at a concentration not exceeding 5 ppm. For peeled potatoes, the maximum permitted wash concentration is 1 ppm. The use of ClO_2 to disinfect fresh-cut fruits and vegetables is not currently permitted in the United States. The occupational exposure limit (ceiling) is 0.1 ppm in air in the United States (instantaneous up to 15 minutes) (OSHA).

Compared to the information that is available on the effectiveness of chlorine as a disinfectant for fruits and vegetables, much less is known about the efficacy of ClO_2. Control of post-harvest fungal pathogens on pears (Spotts and Peters, 1980) and protozoa in water (Chen *et al.*, 1985) has been studied. *In vitro* tests with conidia and sporangiospores of several fungal pathogens of apples and other fruits demonstrated >99% mortality resulting from a 1-minute treatment in water containing 3 or 5 ppm ClO_2 (Roberts and Reymond, 1994). Longer exposure times were necessary to achieve similar mortalities by treatment with 1 ppm. Of the moulds tested, *Botrytis cinerea* and *Penicillium expansum* were least sensitive to ClO_2. Treatment of belts and pads in a commercial apple and pear packing-house with 14-18 ppm ClO_2 in a foam formulation resulted in significantly lower numbers of fungi. It was concluded that ClO_2 has desirable properties as a sanitizing agent for post-harvest decay management when residues of post-harvest fungicides are not desired or not allowed.

The efficacy of ClO_2 in preventing build-up of microorganisms in water for handling cucumbers and on the microorganisms present in fresh cucumbers has been studied (Costilow *et al.*, 1984). At 2.5 ppm, ClO_2 was effective in killing microorganisms in wash-water but, at concentrations up to 105 ppm, failed to reduce the population of microorganisms present in or on fresh cucumbers. It was concluded that many microorganisms were so intimately associated with the cucumber fruit that they were unaffected by chlorine and ClO_2. Reina *et al.* (1995) evaluated the efficacy of ClO_2 in controlling microorganisms in recycled water in a spray-type hydrocooler used to treat pickling cucumbers. Residual ClO_2 at 1.3 ppm was found to optimally control (2-6 $\log_{10}$ CFU/ml reduction) the number of microorganisms in the water. At 0.95 ppm ClO_2, the population was static, while at 2.8 and 5.1 ppm the odour became excessive. Populations of microorganisms on and in cucumbers were not greatly influenced by ClO_2, even at 5.1 ppm. It was concluded that the use of ClO_2 in water used to cool cucumbers seems to be an effective means of controlling microbial build-up, but that it has little effect on the viability of microorganisms on cucumbers.

The effectiveness of ClO_2 in killing *L. monocytogenes* inoculated onto the surface of shredded lettuce and cabbage leaves has been studied (Zhang and Farber, 1996). A 10-minute exposure of lettuce to 5 ppm ClO_2 caused a maximum reduction of 1.1 and 0.8 logs in numbers of *L. monocytogenes* at 4°C and 22°C, respectively, compared with a tap-water control. Similar results were obtained with cabbage. Thus, the maximum reduction in populations of *L. monocytogenes* on shredded lettuce and cabbage treated with ClO_2 at target concentrations up to 5 ppm was only slightly more than 90%, which conforms with observations on the lack of effectiveness of ClO_2 in killing microorganisms on and in cucumbers (Costilow *et al.*, 1984; Reina *et al.*, 1995). As observed with chlorine and other disinfectants, microorganisms differ greatly in their sensitivity to ClO_2, and environmental conditions under which ClO_2 is applied can greatly influence efficacy. The effectiveness of ClO_2 in killing specific pathogenic microorganisms on specific types of fruits and vegetables deserves further research attention.

Bromine

Bromine has had limited use either alone or in combination with chlorine compounds in water treatment programmes, but little is known about its effectiveness as a disinfectant for fruits and vegetables. *Pseudomonas aeruginosa*, a spoilage bacterium, appears to be more resistant to bromine than some other bacteria. Free bromine (200 ppm) does not kill *P. aeruginosa* within 15 minutes at 24°C, but does kill *E. coli, Salmonella* Typhosa and *Staphylococcus aureus* (Gershenfeld and Witlin, 1949). Chlorine is more lethal than dibromodimethyl hydrantoin against *B. cereus* spores (Cousins and Allan, 1967) but is equally effective against *Streptococcus faecalis* (Ortenzio and Stuart, 1964). Others (Kristofferson, 1958; Shere *et al.*, 1962) have observed that the addition of bromine to solutions containing chlorine compounds increases antimicrobial activity, the effectiveness sometimes being synergistic. The occupational permissible exposure limit (PEL) in air in the United States is 0.1 ppm (8-hour time-weighted average) (OSHA).

Iodine

Like chlorine, iodine compounds are widely used for sanitizing food processing equipment and surfaces. Under most conditions, free elemental iodine and hypoiodous acid are believed to be the active antimicrobial agents (Odlaug, 1981). The major iodine formulations are ethanol-iodine solutions, aqueous iodine solutions and iodophors, which are combinations of elemental iodine and nonionic surfactants such as nonyl-phenol ethoxylates (Bartlett and Schmidt, 1957) or a carrier such as polyvinylpyrrolidone (Lacey, 1979). Iodophors have greater solubility in water, are less volatile and irritating to the skin than ethanolic or aqueous solutions of iodine (Lawrence *et al.*, 1957), and have a broad spectrum of activity, including yeasts and moulds. Iodophors are less corrosive than chlorine at low temperatures but vapourize at temperatures above about 50°C where they can be highly corrosive. The efficacy of iodophors is reduced at low temperatures.

Iodophors are most effective at pH 2-5 but can also be active at slightly alkaline pH, depending on other conditions. At concentrations of 6-13 ppm available iodine (pH 6.6-7.0), the time to reduce populations of vegetative bacteria by 90% ranges from 3 to 15 seconds (Gray and Hsu, 1979; Hays *et al.*, 1967; Mosley *et al.*, 1976). Bacterial spores are very resistant to iodine, compared to vegetative cells. Spores of *B. cereus, B. subtilis* and *Clostridium botulinum* (type A) have D values (90% reduction in population) 10-fold to 1000-fold greater than vegetative cells of bacteria treated with 10-100 ppm iodophor (Odlaug, 1981). Although iodophors are minimally affected by organic matter, they may stain equipment used to handle fruits and vegetables and react with starch to form a blue-purple colour. For this reason, the use of iodophors for direct-contact disinfection of many fruits and vegetables has limited potential.

Trisodium phosphate

Trisodium phosphate (TSP) is known to be effective in removing *Salmonella* from poultry (Lillard, 1994) and red meats (Dickson *et al.*, 1994) when applied to carcasses in the form of a chill or rinse-water during processing. Zhuang and Beuchat (1996) investigated the effectiveness of TSP in wash-water in killing *S. Montevideo* on the surface and in core tissue of inoculated mature green tomatoes. Complete inactivation of *Salmonella* (5.18 $\log_{10}$ CFU/cm^2) on the tomato surface was achieved by dipping tomatoes in 15% TSP solution for 15 seconds. Significant (P 0.05) reductions were obtained by dipping tomatoes in a 1% TSP solution for 15 seconds. Populations (5.58 $\log_{10}$ CFU/g) were significantly reduced in core tissue of tomatoes dipped in 4-15% TSP. However, even at 15%, only about a 2 $\log_{10}$ reduction was achieved. It was concluded that the use of TSP as a disinfectant for removal of *Salmonella* from the surface of mature green tomatoes has good potential.

The use of TSP to remove *L. monocytogenes* from shredded lettuce is less promising. Zhang and Farber (1996) reported that treatment of lettuce with 2% TSP had almost no effect on reducing the population of *L. monocytogenes*. Solutions containing more than 10% TSP damaged the sensory quality of lettuce. Other investigators have observed that *L. monocytogenes* is resistant to TSP (Somers *et al.*, 1994). *Escherichia coli* O157:H7, on the contrary, was sensitive to 1% TSP, 10^6 CFU/ml or 10^5 CFU/cm^2 of biofilm being killed within 30 seconds at room temperature or 10°C. *Campylobacter jejuni* was only slightly more resistant than *E. coli* O157:H7. It should be noted that the pH of TSP solutions is in the 11 to 12 range, thus perhaps limiting their application as a disinfectant of fruits and vegetables to commercial use.

Quaternary ammonium compounds

Quaternary ammonium compounds (quats) are cationic surfactants used largely to sanitize floors, walls, drains, and equipment and other food-contact surfaces in fruit and vegetable processing plants. Quats are noncorrosive to metals and stable at high temperature. They are more effective than chlorine against yeasts, moulds and gram-positive microorganisms such as *L. monocytogenes*, and less effective in killing gram-negative bacteria such as coliforms, *Salmonella*, pathogenic *E. coli*, *Pseudomonas* and *Erwinia* species, the latter two being among the major spoilage bacteria of raw vegetables. However, the antimicrobial activity of quats varies considerably, depending on the type used. Because of their surfactant activity, quats have good penetrating ability and appear to form a residual antimicrobial film when applied to most hard surfaces. Quats are relatively stable in the presence of organic matter and properly diluted solutions are odourless and colourless. Their effectiveness is greatest in a pH range of 6-10, thus limiting their potential as disinfectants in highly acidic environments. Quats are not compatible with soaps and anionic detergents. Since most cleaners are anionic, surfaces must be throughly rinsed between cleaning and disinfecting. This group of disinfectants has potential for application to surfaces of uncut fruits and vegetables which subsequently would have their peel, rind or skin removed before consumption.

Acids

Organic acids naturally present in fruits and vegetables or accumulated as a result of fermentation are relied upon to retard the growth of some microorganisms and prevent the growth of others. Foodborne bacteria capable of causing human illness cannot grow at pH values less than about 4.0, so the acidic pH of the edible portions of most fruits precludes them as substrates for proliferation of human pathogens. The pH of many vegetables and a few fruits (e.g. melons), however, is the range at which pathogens can grow.

Some organic acids naturally found in or applied to fruits and vegetables behave primarily as fungistats, while others are more effective at inhibiting bacterial growth. Acetic, citric, succinic, malic, tartaric, benzoic and sorbic acids are the major organic acids that occur naturally in many fruits and vegetables. The mode of action of these acids is attributed to direct pH reduction, depression of the internal pH of microbial cells by ionization of the undissociated acid molecule, or disruption of substrate transport by alteration of cell membrane permeability.

Washes and sprays containing organic acids, particularly lactic acid, have been successfully used to disinfect beef, lamb, pork and poultry carcasses. Application of organic acid washes to the surface of fruits and vegetables for the purpose of reducing populations of microorganisms also has potential. Pathogenic microorganisms on the surface of uncut fruits and vegetables after washing with water or a disinfectant solution, or transferred to the flesh or pulp of fruits or vegetables during the cutting process, can be killed or prevented from growing by applying organic acids. Procedures as simple as applying lemon juice, which contains citric acid as the major acid, to cut fruits have been shown to kill or retard the growth of pathogens. Escartin *et al.* (1989) reported that application of lemon juice to the surface of papaya (pH 5.69) and jicama (pH 5.97) cubes inoculated with *Salmonella* Typhi reduced populations compared to the control, but growth resumed after several hours. Survival of *C. jejuni* inoculated onto watermelon (pH 5.5) and papaya (pH 5.6) cubes as affected by treatment with lemon juice was investigated by Castillo and Escartin (1994). The amount surviving 6 hours after treatment ranged from 7.7% to 61.8% in fruits not treated with lemon juice acid and from 0% to 14.3% in fruits with lemon juice added. Treatment appeared to be more effective in killing *Campylobacter* on papaya than on watermelon, perhaps because of differences in porosity or buffer capacity of these fruits.

Shapiro and Holder (1960) observed that treatment of salad vegetables with up to 1500 ppm citric acid did not affect bacterial growth during a subsequent 4-day storage period at 10°C. Treatment with 1500 ppm tartaric acid reduced total counts 10-fold. Treatment of cut lettuce, endive, carrots, celery, radishes and green onions with 2000 ppm sorbate or 10,000 ppm ascorbic acid, alone and in combination, resulted in less than 1 log difference in populations of aerobic microorganisms after 10 days of storage in 4.4°C (Priepke *et al.*, 1976).

Removal of pathogenic bacteria from leafy salad greens by treating with acetic acid has been studied. Reduction in counts of *Yersinia enterocolitica* inoculated onto parsley leaves from 10^7 CFU/g to <1 CFU/g by washing in a solution containing 2% acetic acid or 40% vinegar for 15 minutes was achieved by Karapinar and Gonul (1992). No viable aerobic bacteria were recovered after a 30-minute dip in 5% acetic acid, whereas a vinegar dip resulted in a 3-6 $\log_{10}$ decrease in the number of aerobic bacteria, depending on vinegar concentration and holding time.

The effects of lactic and acetic acid, either alone or in combination with chlorine, on survival of *L. monocytogenes* inoculated onto shredded lettuce were studied by Zhang and Farber (1996). Compared to lettuce washed in tap-water, only 1% lactic acid and combinations of 0.5% or 1% lactic acid and 100 ppm chlorine reduced numbers of *L. monocytogenes*. Lactic acid (0.75% or 1%) in combination with 100 ppm chlorine was more effective in reducing levels of *L. monocytogenes* than were either lactic acid or chlorine alone. Results obtained with acetic acid were similar to those with lactic acid.

Treatment of ready-to-use salads with 90 ppm peracetic acid has been shown to reduce total counts and faecal coliforms by nearly 100-fold, similar to reductions with 100 ppm chlorine

(Masson, 1990). Reduced growth of microflora during subsequent storage was attributed to residual effects of acetic acid released by degradation of peracetic acid. Peroxyacetic acid has been used extensively as a sanitizer for food processing equipment where it is particularly effective against biofilms. It also holds potential as a disinfectant for fruits and vegetables, although its efficacy in killing infectious microorganisms attached to the surface of plant materials has not been extensively studied.

The observation that washing or rinsing fruits and vegetables with organic acids will reduce populations of some types of pathogenic bacteria would indicate that applying vinegar or lemon juice holds promise as a simple and inexpensive household disinfection procedure. Indeed, such treatments are likely to reduce the risk of illness associated with potentially contaminated fruits and vegetables. A possible disadvantage is that these treatments may change the flavour and aroma of treated products.

Hydrogen peroxide

Hydrogen peroxide (H_2O_2) can have a lethal or inhibitory effect on microorganisms, depending on the pH, temperature and other environmental factors (Juven and Pierson, 1996). Saper (1996) has studied the efficacy of H_2O_2 in improving the microbiological quality and extending the shelf-life of minimally processed fruit and vegetable products. Hydrogen peroxide vapour treatments were highly effective in reducing microbial numbers on whole cantaloupes, grapes, prunes, raisins, walnuts and pistachios. However, similar treatment induced browning in mushrooms exposed to levels necessary to delay spoilage by *Pseudomonas tolaasii*. Exposure to H_2O_2 vapour caused bleaching of anthocyanins in strawberries and raspberries. Dipping freshly-cut green bell pepper, cucumber, zucchini, cantaloupe and honeydew melon in H_2O_2 solution had no adverse effect on appearance, flavour or texture, but it induced severe browning of shredded lettuce. Dip treatment significantly reduced the population of *Pseudomonas* on these products but had no measurable effect on yeasts and moulds.

Reductions in populations of *Salmonella* on alfalfa sprouts are similar by dipping sprouts in 200 and 500 ppm chlorine or 2% and 5% H_2O_2, respectively, for 2 minutes (Beuchat, 1997). Slightly more than 2 $\log_{10}$ CFU/g reduction was observed after treatment with 200 ppm chlorine or 2% H_2O_2. The effectiveness of the same concentrations of chlorine and H_2O_2 in eliminating *Salmonella* from cantaloupe cubes was much less, however, with reductions of less than 1 $\log_{10}$ CFU/g. Alfalfa sprouts and, to a lesser extent, cantaloupe cubes took on a mildly bleached appearance after treatment with 5% H_2O_2. Results from studies on a limited number of fruits and vegetables indicate that H_2O_2 has potential for use as a disinfectant. Some fruits and vegetables (e.g. mushrooms, some types of berries and lettuce), however, may not be amenable to disinfecting with H_2O_2 because of adverse changes in surface colour. Additional work to determine the effectiveness of H_2O_2 in killing pathogens on a wide range of raw fruits and vegetables is warranted. The occupational PEL in air in the United States is 1 ppm (8-hour time-weighted average) (OSHA).

Ozone

Treatment of drinking-water with ozone for the purpose of killing microorganisms has been practised for nearly a century. *Salmonella* Typhimurium, *Y. enterocolitica, S. aureus,* and *L. monocytogenes* are among the pathogens sensitive to treatment in ozonated (20 ppm) water (Restaino *et al.*, 1995). Enteric viruses (Finch and Fairbairn, 1991) and oocysts of protozoa such as

Cryptosporidium parvum (Korich *et al.*, 1989) are also sensitive to ozone. Greater than 90% inactivation of *C. parvum* was achieved by treatment with 1 ppm ozone for 5 minutes (Peeters *et al.*, 1990). *C. parvum* oocysts are about 30 times more resistant to ozone and 14 times more resistant to ClO_2 than *Giardia* cysts under the same conditions.

The use of ozone to disinfect various types of foods has been investigated. Preservation of fish (Haraguchi *et al.*, 1969), reduction of aflatoxin in peanuts and cottonseed meals (Dwankanath *et al.*, 1968), reduction of microbial populations on poultry (Sheldon and Brown, 1986), and reduction of microbial populations on bacon, beef, butter, cheese, eggs, mushrooms, potatoes and fruits (Gammon and Kerelak, 1973; Kaess and Weidemann, 1968) using gaseous ozone have been studied. High relative humidity or aqueous conditions generally favour microbicidal activity. The lethal effect of ozone is a consequence of its strong oxidizing power. For this reason, physiological injury of produce, for example in bananas, can result from exposure to concentrations as low as 1.5 ppm without damage to sensory qualities (Horvath *et al.*, 1985). After eight days, black spots appear on the skin of bananas if the ozone concentration is maintained at 25-30 ppm. Extension of shelf-life of oranges, strawberries, raspberries, grapes, apples and pears can be achieved by treatment with ozonated water. In addition to the antimicrobial effects of ozone, oxidation of ethylene also occurs, thus retarding metabolic processes associated with ripening. Concentrations in the range of 2-3 ppm for berry fruits and 40 ppm for oranges greatly reduce microbial populations.

Because of its instability, ozone must be generated at the usage site. Users should also be aware that, because of the strong oxidizing power of ozone, metal and other types of surfaces with which it comes into contact are subject to corrosion or other deterioration. The rate of deterioration depends on the concentration of ozone. Nevertheless, the use of ozonated wash and flume-waters in fruit and vegetable handling and processing operations provides a method to control build-up of microbial numbers, particularly in recycled water, and deserves consideration for routine use as a disinfectant for fruits and vegetables. The occupational PEL in air in the United States is 0.1 ppm (8-hour time-weighted average) (OSHA).

Irradiation

The application of ionizing irradiation (e.g. gamma rays from ^{60}Co or ^{137}Cs) to raw fruits and vegetables is a means of extending shelf-life (Thayer *et al.*, 1996). Decay caused by indigenous microflora and post-handling contaminants can be eliminated or delayed by dose levels that do not adversely affect the sensory qualities of many fruits and vegetables. Dose levels of 1 to 3 kGy, depending upon the type of fruit or vegetable, are sufficient to kill large numbers of most moulds, yeasts and bacteria naturally present on produce as it is taken from the field (Farkas, 1997; Moy, 1983; Urbain, 1986).

Factors influencing lethality include the level of sensitivity of the target insect or microorganism(s) to irradiation, post-harvest and pretreatment conditions of fruits and vegetables, and environmental conditions (e.g. relative humidity and temperature) surrounding fruits and vegetables at the time of treatment. Extensive studies to determine the efficacy of irradiation treatment in disinfestation (CAST, 1986; 1989) and abating post-harvest fungal diseases have been reviewed (Clarke, 1959; Willison, 1963; Moy, 1983; Wilkinson and Gould, 1996). *Botrytis, Rhizopus* and *Mucor* species are among the major spoilage moulds of raw fruits and vegetables, and are inactivated by dose levels of 1.2 to 3.0 kGy. A dose of 3 kGy, for example, will extend the shelf-life of strawberries stored at 5°C by about 1 week (Sommer and Maxie, 1966). Guidelines for disinfestation and quarantine treatment of raw fruits and vegetables have been issued (ICGFI, 1991a; 1991b; ASTM, 1993).

The effectiveness of ionizing irradiation in killing microorganisms capable of causing human illness has been studied but most investigations have been done using *in vitro* conditions or foods of animal origin (Farkas, 1989; 1997; Monk *et al.*, 1995; Mossel and Stegeman, 1985). Vegetative cells of pathogenic bacteria are sensitive to irradiation, D_{10} values being in the range of 0.2-0.8 kGy, depending on environmental conditions. Foodborne parasites are also generally sensitive to irradiation. With the exception of some nematodes such as *Aniostrongylus cantonensis*, *Gnatostoma spinigerum* and *Anisakis* sp., a dose below 1 kGy is sufficient to control infection caused by protozoa, trematodes and cestodes (Loaharanu and Murrell 1994). Higher doses are required, however, for inactivation of viruses. D_{10} values for inactivating viruses (Mallett *et al.*, 1991; Sullivan *et al.*, 1973) may be more than 10-fold higher. Therefore, irradiation cannot be used for inactivation of viruses. Nevertheless, ionizing irradiation could be an extremely effective tool in reducing populations of pathogenic microorganisms and parasites from the surface of raw fruits and vegetables. Its application on a large scale for the purpose of reducing the risk of disease associated with the consumption of potentially contaminated raw fruits and vegetables would appear to have exceptional merit. There is a need, however, to evaluate the tolerance of most fruits and vegetables to the radiation doses required for controlling various pathogenic microorganisms.

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Conclusion

Several pathogenic bacteria, viruses and parasites capable of causing human disease can be found on raw fruits and vegetables. Some of these microorganisms are capable of growing on whole, minimally processed or cut fruits and vegetables under routine handling and storage conditions. It is essential that interventions be made to prevent contamination of raw fruits and vegetables and, failing this, to remove disease-causing microorganisms prior to consumption. However, none of the chemical or physical treatments currently used to disinfect raw fruits and vegetables can be relied on to eliminate all types of pathogens from the surface or internal tissues when using application conditions that will not adversely affect sensory or nutritional qualities. Irradiation is a powerful tool against bacteria and parasites but cannot be relied on to eliminate viruses.

The efficacy of various disinfectants and sanitizing methods for reducing populations of microorganisms on raw fruits and vegetables varies greatly. Differences in surface characteristics of fruits and vegetables, type and physiological state of microbial cells, and environmental stress conditions interact to influence the activity of disinfectants and sanitizers. Conditions under which chlorine is effective in killing microorganisms on fruits and vegetables have been studied most extensively. Nevertheless, its performance in relation to other chemical and physical treatments under various conditions of application has not been clearly defined. Although there is a lack of extensive scientific data from which to extract firm conclusions concerning the efficacy of disinfectants for raw fruits and vegetables, some general conclusions can be drawn:

- Efficacy of disinfectants varies with different fruits and vegetables, characteristics of their surfaces, temperature and type of pathogen.
- *Listeria monocytogenes* is generally more resistant to disinfectants than *Salmonella, Escherichia coli* O157:H7 and *Shigella*.
- Little is known about the efficacy of disinfectants in killing parasites and viruses on fruits and vegetables.
- Washing fruits and vegetables in potable water removes a portion of microbial cells. In some instances, vigorous washing can be as effective as treatment with water containing 200 ppm chlorine, which generally reduces populations by 10-100-fold.
- Heavily contaminated fruits and vegetables should be subjected to a double wash treatment. Success in removing soil or faecal matter, and the contaminants therein, is more likely to be achieved by first washing in potable water and then washing or rinsing in water containing a disinfectant.
- The temperature of wash-water should be higher than that of the fruits or vegetables in order to minimize uptake of microorganisms by tissues.
- The lethal effect of chlorine occurs within the first few seconds of treatment. The population of microorganisms decreases as the concentration of chlorine increases to about 300 ppm, above which effectiveness is not proportional to increased concentration.
- Leaving fruits and vegetables wet after disinfecting or washing can negate any beneficial effect of treatment.
- Chlorine dioxide is useful in controlling populations of microorganisms in wash-water but varies in efficacy in killing microorganisms on the surface of fruits and vegetables.

- Bromine and iodine may have limited potential as disinfectants for fruits and vegetables, partly because of their adverse effect on sensory quality.
- Trisodium phosphate has good potential as a disinfectant for whole fruits and vegetables in a commercial setting. Use in households may be limited, however, because the high alkalinity of TSP may cause skin irritation.
- Although disinfectants have variable effects on pathogen control on fresh fruits and vegetables, they are certainly useful for sanitizing wash-water to prevent contamination of the produce that could result from using waters that are not microbiologically safe.
- Organic acids (e.g. acetic, lactic, citric and peroxyacetic acids) have good potential as disinfectants for fruits and vegetables, but conditions under which they are most effective have not been defined.
- Ozonation of wash-water reduces numbers of microorganisms, thus resulting in reduced numbers on the surfaces of fruits and vegetables.
- Faecal matter or water containing faeces should never come into contact with fruits and vegetables, as even the most powerful treatment (e.g. irradiation) cannot be relied on to eliminate some of the pathogens they may contain.
- Prevention of contamination of fruits and vegetables with pathogens at all points from the field to the plate, through application of good agricultural practices (GAP), good manufacturing practices (GMP) and HACCP programmes, is preferred to application of chemical disinfectants after contamination has occurred.

There is a lack of information on the extent and type of microbial contamination of raw fruits and vegetables on an international scale. Nevertheless, observations support these conclusions, and can serve as a basis for developing recommendations and plans of action.

Future directions and recommendations

Application of the principles inherent in or deriving from the following recommendations will result in reduced risk of illness associated with raw fruits and vegetables.

1. Further basic and applied research is needed in order:
   - to better understand modes of contamination of raw fruits and vegetables before harvesting, when subjected to various agronomic practices, and during post-harvest handling;
   - to determine conditions that influence attachment, growth, survival and death of human pathogenic microorganisms on fruits and vegetables;
   - to establish and validate disinfection methods, individually and in combination, that minimize risk of illness caused by microorganisms on fruits, vegetables and other plant materials eaten raw (such research should also address the short-term and long-term toxicity of residues or reaction products resulting from chemical or physical treatments);
   - to develop highly effective treatments for removing pathogens from a wide range of raw produce (a single type of treatment will not be suitable for all fruits and vegetables. Caution must be taken in the development of sanitizing or disinfecting treatments so that degradation of nutritional components of fruits and vegetables is minimal and the formation of compounds with potential toxic or carcinogenic properties is avoided);
   - to develop or improve analytical techniques to detect low numbers of microorganisms, particularly viruses and parasites, on or in raw fruits, vegetables and plant materials before and after treatment with various disinfectants.

2. Control of pathogenic microorganisms should involve multidisciplinary teams with a wide range of technical, sociological, educational and administrative skills. Health education programmes should fully describe the impact of infections that can be associated with raw fruits and vegetables.

3. Hygienic principles should be applied from production to consumption of fruits and vegetables (agriculture, transport, manufacturing and processing, distribution, and preparation for consumption). Disinfection should be applied where appropriate. However, interventions to minimize contamination of fruits and vegetables at any point in the chain should be preferred to remedial or corrective action.

4. Effective surveillance programmes should be established to determine the presence and prevalence of specific pathogenic bacteria, viruses and parasites on raw fruits and vegetables at the point of importation.

5. Epidemiological studies should be carried out to address the role of raw fruits and vegetables as vehicles of microorganisms capable of causing human diseases.

6. There should be continual training of fruit and vegetable growers and handlers at all levels in order to control microbiological hazards that may be influenced by current and changing aquacultural, agronomic, processing, distribution and preparation practices.

7. Conferences, workshops and symposia specifically addressing the control of human infections associated with the consumption of contaminated raw fruits and vegetables should be held in places where they will have greatest impact.

8. Professional and domestic food handlers and consumers should be better educated about the principles of personal hygiene and decontamination of raw fruits, vegetables and plant materials. Changes in risk of illness related to ageing and health status of consumers should be more clearly defined.

9. Quantitative microbiological risk assessment of various human infections and intoxications that can be linked to the consumption of contaminated raw fruits, vegetables and plant materials should be undertaken.

10. Cost-benefit and environmental impact assessments should be made for each method of disinfection.

11. The establishment of effective GAP, GMP and HACCP programmes that would cover all aspects of growing, harvesting, packing, transport, processing, distribution, and preparation of raw fruits and vegetables is strongly encouraged. Since contamination of raw produce with pathogens can occur at any point in the system, all stages must be considered when devising HACCP programmes. Assistance from, and collaboration between, academic institutions, public health authorities, food control agencies, trade organizations and the private sector in developing HACCP programmes for the raw fruit and vegetable industry will be necessary to minimize the risk of disease. An integral part of this process will involve identifying critical control points at which measures should be taken to prevent contamination and to decontaminate. In the interim, where lack of science-based information prevents identification of critical control points for specific segments of the fruit or vegetable growing and handling chain, this should be acknowledged and GAP and GMP established. The development of HACCP programmes will eventually follow.

12. Several diseases caused by viruses, parasites and some types of pathogenic bacteria that are linked to the consumption of raw fruits and vegetables are transmitted via the faecal-oral route. It

is imperative that individuals handling raw produce at every stage, from the field to the point of consumption, have an understanding of hygiene sufficient to prevent contamination. Application of improperly composted manure or water containing raw sewage to fields, or the use of water contaminated with faeces during processing of fruits and vegetables, must be avoided. These responsibilities must be shared by the grower as well as the processor, shipper, distributors and food handlers. Training of food handlers at all level of the food chain, as well as education of the consumer, are key elements in a total system approach to reducing the risk of produce-borne illnesses.

13. Finally, but not least in importance, there is a need to further develop and enhance international epidemiological surveillance programmes for foodborne diseases. Collaboration in such programmes, whether between countries or continents, will generate information that will be useful in developing guidelines for reducing the risk of disease that may be caused by raw fruits and vegetables.

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Surface decontamination of fruits and vegetables eaten raw: a review

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Contents

Preface

Prevention of contamination is the most efficient way to ensure food safety and prevent foodborne illness. Thus, every effort should be made to protect food from primary sources of contamination. This is, however, not always possible. Raw foodstuffs, particularly fruits and vegetables grown close to the soil, may be contaminated with various foodborne pathogens.

In some countries, chemical disinfectant agents are used to decontaminate the surface of fruits and vegetables, in addition to washing with water. It is not clearly known to which extent these agents are effective, what their optimum conditions of use are, and whether they have adverse toxicological effects.

The present document attempts to provide an insight into some of these concerns. It presents a review of pathogens associated with fruits and vegetables and a literature review of studies carried out to investigate the efficacy of a range of methods used for surface decontamination. However, it does not review the potential adverse effects of using chemical disinfectants. Further work is needed on this subject. Therefore, the document should not be seen as a recommendation of the use of chemical disinfectant agents for surface decontamination of fruits and vegetables.

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Summary

Outbreaks of human diseases associated with the consumption of raw fruits and vegetables often occur in developing countries and have become more frequent in developed countries over the past decade. Factors thought to influence the occurrence and epidemiology of these diseases include irrigation and other agronomic practices, the general level of hygiene in handling fruits and vegetables, international travel, globalization of the supply and distribution of raw produce, the introduction of pathogens into new geographical areas, changes in the virulence and environmental resistance of pathogens, decrease in immunity among certain parts of the population (particularly the elderly), and changes in eating habits.

Fruits and vegetables eaten raw, as well as food of animal origin, have long been known to serve as vehicles for transmission of infectious microorganisms in developing countries. The number of confirmed cases of illness associated with raw fruits and vegetables in industrialized countries, on the other hand, has been relatively low compared to the number of recorded cases due to foods of animal origin. The impact of agronomic practices, handling, processing, distribution and marketing on risks associated with microbiological hazards of raw fruits and vegetables is not well understood.

This document reviews the health hazards associated with fruits and vegetables and the effects of different decontamination methods in eliminating or reducing the microbial load. Disinfectants are effective in reducing the bacterial load but their efficacy depends on the types of fruits and vegetables and characteristics of their surface, the method and the procedure used for disinfection (e.g. temperature, pH etc), and on the type of pathogen. *Listeria monocytogenes* is generally more resistant to disinfectants than *Salmonella*, pathogenic *Escherichia coli* and *Shigella*, but little is known about the efficacy of disinfectants in killing parasites and viruses on fruits and vegetables. Vigorously washing fruits and vegetables with safe water reduces the number of microorganisms by 10-100-fold and is often as effective as treatment with 200 ppm chlorine. Treatments with chlorine dioxide, trisodium phosphate, organic acids or ozone have potential for removing pathogenic microorganisms from raw fruits and vegetables. When applicable, food irradiation is one of the most powerful methods of decontamination.

Prevention of contamination at all points of the food chain from primary production to the final consumer is preferred over the application of disinfectant after contamination occurs. Prevention can be achieved through application of the principles of food hygiene and the Hazard Analysis and Critical Control Point system (HACCP).

Research is needed to better understand not only the mechanisms through which pathogens can contaminate raw fruits and vegetables but also the procedures for killing or removing pathogens once they are present, either on the surface or in internal tissues, and the analytical methods for their detection. A quantitative microbiological risk assessment should be undertaken of human infections and intoxications that can be linked to the consumption of contaminated raw fruits, vegetables and plant materials. The development of a highly efficient, international epidemiological surveillance system for better understanding the role of raw fruits and vegetables as vehicles for infectious diseases is critical. Information generated by such a system would be valuable in establishing more meaningful practices and guidelines for preventing contamination and for decontaminating raw fruits and vegetables.

Continual training of fruit and vegetable growers and handlers at all levels should be carried out in order to control microbiological hazards that may be influenced by current and changing practices in aquaculture, agronomy, processing, marketing and preparation. The use of water and fertilizers free of pathogenic microorganisms at all stages of growing and handling and, just as importantly, an appropriate level of understanding of the importance of personal hygiene by everyone involved is essential if the risk of illness is to be minimized. Regardless of the stringency of hygienic measures at production and processing stages of the food chain, consumers should be educated to realize the importance of adequate washing of fruits and vegetables.