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Erratum

Review
Microbiological safety evaluations and recommendations on fresh produce by C. De Roever. *Food Control*, 9, 321–347 (1998)☆

The publisher regrets that the above paper was published without corrections, leading to many errors appearing in the published version. The corrected version is reproduced below.

Microbiological safety evaluations and recommendations on fresh produce

National Advisory Committee on Microbiological Criteria for Foods

Washington, USA

Abstract

In 1995, the National Advisory Committee on Microbiological Criteria for Foods (Committee) was asked to investigate and characterize the association between cases for foodborne illness and fresh produce. The Committee was asked to provide recommendations that could be employed to reduce the risk of foodborne outbreaks associated with these commodities. In furtherance of this assignment, the Committee reviewed current epidemiologic data, the microbial ecology of the outbreak-associated organisms, and considered the current industry practices used for growing, harvesting, packing and distribution. An evaluation of these data provided a basis for hazard identification and the development of related control measures.

The Committee has developed seven specific recommendations: Good Agricultural Practices (GAP) and Good Manufacturing Practices (GMP) should be developed that will provide guidance on those agricultural and processing steps that can reduce pathogen levels on fresh produce. While Hazard Analysis Critical Control Point Programs (HACCP) would be likely to provide the greatest assurance of the safety of these products, there are presently insufficient data upon which to develop such progams. Proactive and practical education programs are needed at all steps in the process, i.e., from the field to the consumer's plate. Additional data are needed to conduct effective risk assessments of the microbial hazards associated with fresh produce. Better product identification and tracing systems are needed for outbreak investigations. Research must be conducted to fill some of the current knowledge gaps in order that improved intervention strategies can be employed. Steps should be taken to 'streamline' approval processes for new technologies that will reduce/eliminate microbial hazards.

Nomenclature

CCP	Critical Control Point
CDC	Centers for Disease Control and Prevention
ETEC	Enterotoxigenic *Escherichia coli*
EPA	Environmental Protection Agency
FDA	Food and Drug Administration
FFVA	Florida Fruit and Vegetable Association
GAP	Good Agricultural Practices
GMP	Good Manufacturing Practices
HACCP	Hazard Analysis and Critical Control Point
HUS	Hemolytic Uremic Syndrome
IFPA	International Fresh-cut Produce Association
NACMCF	National Advisory Committee on Microbiological Criteria for Foods

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PFGE	Pulsed-Field Gel Electrophoresis
QSR	Quick Service Restaurant
SSOP	Sanitation Standard Operating Procedure
United	United Fresh Fruit and Vegetable Association
USDA	US Department of Agriculture
WGA	Western Growers Association

1. Introduction

In the past two decades, there has been a noticeable increase in the consumption of fresh fruits and vegetables in the United States and a marked increase in the global distribution of produce. An abundance of research over the last decade has shown that diets which are low in fat and high in fiber, and that include liberal consumption of fruits and vegetables, are protective against many cancers and lessen the risk of coronary heart disease. Unfortunately, too many Americans fail to consume balanced diets and nearly two-thirds of all cancers are attributed to dietary behavior. Consequently, changing dietary behavior represents one of today's most important public health challenges (Kennedy et al., 1996; Kushi et al., 1995; National Research Council, 1989).

Government and independent nutrition and health authorities agree that Americans should increase their consumption of fruits and vegetables to at least five servings a day (Kennedy et al., 1996; Kushi et al., 1995). Many elements of federal policy reflect this view, and the aim of increasing the consumption of fruits and vegetables is a core principle of such initiatives as the *U.S. Dietary Guidelines, Healthy People 2000*, and the National Cancer Institute's *Five A Day Program*.

However, public health officials have documented significant increases in the number of produce-associated foodborne disease outbreaks in the US. According to the Centers for Disease Control and Prevention (CDC), the number of reported produce-related outbreaks per year doubled between the period 1973–1987 and 1988–1992 when illness due to botulism and mushroom and 'salad'; (which included salads containing non-produce items) associated illness were excluded. Substantial increases in produce-related human illness were also observed in 1995. Outbreak data linked *Salmonella* Stanley with alfalfa sprouts, *Salmonella* Hartford with orange juice, *Shigella* spp. with lettuce and scallions, *Escherichia coli* O157:H7 with lettuce varieties, and hepatitis A virus with tomatoes. In the past two years, there have been additional outbreaks linking *Cryptosporidium* with unpasteurized apple cider, *Cyclospora* with raspberries, mesclun lettuce, and basil/basil-containing products, *E. coli* O157:H7 with unpasteurized apple cider, and hepatitis A virus in sliced, sugared and frozen strawberries. These are only limited examples of identified outbreaks associated with fresh and minimally processed produce.

In response to this growing concern about fruits and vegetables as a source of infectious foodborne disease, in 1995, the Food and Drug Administration (FDA) asked the National Advisory Committee on Microbiological Criteria for Foods (NACMCF) to investigate and characterize the association of foodborne disease and microbial pathogens with fresh produce. The Fresh Produce Subcommittee was formed and has documented the relevant epidemiology, microbial ecology of organisms associated with produce outbreaks, and has reviewed current industry practices and initiatives related to the growing, harvesting, packing and distribution of produce. The subcommittee evaluated data to identify hazards and related controls, discussed research needs, and provided recommendations to prevent future produce-associated illnesses.

The Subcommittee also toured harvesting, processing, and packing facilities for fresh produce in south central Florida to get a better understanding of current industry practices. Producers in the area and their trade associations have been very helpful and interested in working with the Subcommittee to address the potential microbiological hazards associated with fresh produce.

There are many questions about the transmission of microorganisms from their potential reservoirs to fruits and vegetables, and any vectors which may be involved in this process. While all produce items have factors in common, it is important to recognize that each fruit and vegetable has a unique combination of composition and physical characteristics, growing and harvesting practices, cooling techniques, and optimum storage temperatures and environment. For example, berry fruits, like strawberries and raspberries, are very delicate and highly perishable and are not washed after harvest, tomatoes are usually picked in the mature green stage, flushed from large field totes with water, flumed into the packing house, and after inspection and grading, packed into boxes. Apples have a much smoother skin and more delicate texture than citrus fruits which have a relatively hard and rough skin. All of these characteristics must be taken into consideration when considering potential microbiological hazards and controls.

In addition to whole produce, this document considers processed products, including minimally processed produce, and unpasteurized juices. The committee recognizes that different risk characteristics exist for these products because the protective surface integrity of the whole product has been broken. In the majority of the outbreaks associated with fresh produce, minimally processed products, and unpasteurized

products, the source of contamination remains unknown. Therefore, the inclusion of minimally processed products and unpasteurized juices, which are made from whole produce, does not implicate the whole commodity as the source of contamination.

This report constitutes the Committee findings on the association of foodborne disease and microbial pathogens with fresh produce, along with strategies for their control.

2. Epidemiology of foodborne outbreaks related to fresh produce

2.1. Introduction

In recent years, fresh produce has been determined to be the vehicle of transmission in several outbreaks. This experience has focused attention on the question of whether and to what extent produce-associated illness is a public health problem.

A logical place to look for the answer to this question is in the national database on the epidemiology of foodborne disease maintained by CDC. These data are derived from four major sources: epidemic investigations, an ongoing outbreak surveillance system specific for foodborne disease, laboratory-based surveillance, and in the past 2 years, an active surveillance system for several foodborne pathogens. Each system has characteristics that limit the inferences that can be drawn from the data.

Of these four sources of data, the outbreak surveillance system for foodborne disease has the most longitudinal experience with the largest scope and this could be expected to show the extent to which illness transmitted by fresh produce is occurring. However, for outbreak recognition and reporting, particular characteristics of the food commodity, the pathogen and the resulting illness, make it more likely for the outbreak to be recognized, investigated and reported. These characteristics include: (1) illness with a distinctive symptom complex serious enough to warrant medical attention, which has a short incubation period and which is a notifiable disease; (2) a pathogen with established methods of detection in clinical specimens and food samples, which has already been recognized to cause foodborne disease, and for which diagnostic services are readily available; and (3) a product which has an extended shelf life or is frozen (and thus still available for testing), which has limited distribution and is uncommonly consumed. Outbreaks due to meat and poultry have many of these characteristics, whereas those due to fresh produce do not. For these reasons it is likely that many outbreaks from fresh produce have been unrecognized and unreported.

2.2. The importance of fresh produce as a vehicle for foodborne pathogens

Data from the CDC foodborne disease outbreak surveillance system from 1973 to 1992 suggest at least a doubling in the annual number of reported produce-associated outbreaks, the number of persons affected annually in those outbreaks and in the proportion of outbreaks due to fresh produce among those with an identified food vehicle.

The CDC's national database on foodborne disease identifies a number of pathogen/produce combinations as sources of foodborne disease. Inherent limitations in the database do not allow it to be used in a quantitative manner to define the magnitude or scope of the problem or to provide precise calculations on changes in secular trends over time. However surveillance data from Minnesota (Appendix A, Table 7 and Table 8) provide a more detailed look at the problem and corroborate the observation that fresh produce is emerging as a significant commodity group which can and does transmit pathogens.

2.3. Pathogens epidemiologically implicated in fresh produce-associated diseases

Many different bacteria, viruses, and protozoa have been linked epidemiologically to fresh produce-associated diseases. Appendix A (Table 6) shows the etiology of fresh produce-associated outbreaks reported to CDC in 1973–87 and 1988–92. In both time periods, the etiologic agent was unknown in more than 50% of outbreaks.

Most oubreaks with identified etiology were of bacterial origin; *Salmonella* spp. was most commonly reported in both time periods. However, the number of outbreaks caused by hepatitis A virus has increased, and diagnostic difficulties make it likely that outbreaks due to Norwalk-like viruses are under-recognized and therefore under-reported. Table 1 lists the pathogens most commonly associated with foodborne disease that were due to fresh produce and the food items that have been linked epidemiologically to outbreaks caused by these pathogens. Most pathogens have been linked to several types of produce. Therefore, many possible combinations of pathogen/food item exist for fresh produce-associated diseases.

2.4. Why is there an increase in foodborne diseases associated with fresh produce?

A number of possible explanations exist for the increase in reported produce-associated outbreaks of foodborne disease. Some factors could cause an artificial increase in reported incidence, and these must be ruled out before concluding that the reported increase is real.

Table 1
A literature review of pathogens causing outbreaks associated with fresh or frozen produce.

Pathogen	Produce	Reference
Shigella spp.	Lettuce	Davis et al. 1988; Frost et al., 1995; Kapperund et al., 1995; Martin et al., 1986
	Green onions	Cook et al. 1995
Salmonella spp.	Sliced tomatoes	Wood et al. 1991; CDC, 1993
	Sprouts	O'Mahony et al. 1990; Mahon et al. 1997; Van Beneden, 1996; CDC, 1997g
	Sliced watermelon	Gayler et al. 1955; CDC, 1979; Blostein, 1993
	Sliced cantaloupe	Ries et al. 1990; CDC, 1991b
	Unpasteurized orange juice	CDC, 1995c; Parish, 1997
Escherichia coli O157:H7	Unpasteurized apple cider/juice	Besser et al. 1993; CDC, 1996c; CDC, 1997a
	Lettuce varieties	Anonymous, 1995; CDC, 1995a; CDC, 1995b;
	Alfalfa sprouts	CDC, 1997g
Enterotoxigenic *E. coli* (ETEC)	Carrots	CDC, 1994
Vibrio cholerae	Coconut milk	CDC, 1991a
Listeria monocytogenes	Cabbage	Schlech et al., 1983
Bacillus cereus	Sprouts	Portnoy et al., 1976
Hepatitis A virus	Lettuce	Rosenblum et al. 1990
	Raspberries	Reid and Robinson, 1987
	Frozen strawberries	Niu et al. 1992; CDC, 1997b
	Sliced tomatoes	Williams et al., 1995
Norwalk/Norwalk-like virus	Sliced melon	Iversen et al., 1987
	Green salad	Griffin et al., 1982
	Celery	Warner et al., 1991
Cyclospora cayetanensis	Raspberries	CDC, 1996a; CDC, 1996b; CDC, 1997c; CDC, 1997d
	Mesclun lettuce	CDC, 1997e
	Basil/basil-containing products	CDC, 1997f
Cryptosporidium parvum	Unpasteurized apple cider	Millard et al. 1994; CDC, 1997a

For example, improved diagnostics and amplified surveillance efforts could both stimulate case ascertainment, but these causes would not be expected to selectively increase reporting of produce-associated outbreaks. If general awareness of the potential role of produce in disease transmission had recently increased, this could cause an artificial increase in reported cases. This factor could explain part of the observed increase by shifting outbreaks that would have been reported as having an unknown source into the produce category. Questions about produce consumption have been routintely asked for decades but have only recently identified large numbers of outbreaks. Therefore, it is likely that the observed increase in reported produce-associated outbreaks reflects an actual increase.

Many factors may be involved in the changing epidemiology of produce-associated disease. Changing food industry practices and demographics may influence the epidemiology of produce-associated disease. For example, larger and more centralized production units and a longer food chain could permit amplification of pathogen numbers and their distribution to large numbers of persons in geographically dispersed areas. The increase in global trade makes food from around the world available to the average customer and brings an end to seasonality in the availability of fresh produce, but also potentially exposes consumers to exotic microflora. Consumer demand for convenience has increased consumption of produce that has undergone some kind of minimal, non-thermal processing, e.g., fresh-cut fruits and fresh-squeezed juice. When this handling is followed by time/temperature combinations that permit pathogens to survive and grow, the risk of foodborne illness increases. In addition, changing social demographics are increasing the proportion of the population that is elderly, immunocompromised, or suffering from chronic diseases. These factors increase the population at highest risk for foodborne infections.

Changes in consumer food preferences have resulted in an increase in the consumption of fresh produce (e.g., a 27% increase in fresh produce consumption in the US from 1970 to 1993), which has increased the number of persons exposed to produce-associated pathogens. Another changing food consumption practice is increased use of salad bars and the number of meals being eaten outside the home, increasing the risk of food handling errors with fresh produce. Many consumers also prefer 'natural' and 'organically' cultivated produce, which could result in the increased use of manure rather than chemical fertilizers in food production. Improperly treated manure may contain enteric pathogens such as *Salmonella* spp. and *E. coli* O157:H7.

2.5. Investigation of foodborne outbreaks associated with fresh produce

The investigation of fresh produce-associated outbreaks is usually difficult. Distribution patterns for fresh produce in the US generally make lots widely dispersed, and contamination of produce generally is sporadic and low-level. Therefore, fresh produce-associated outbreaks are typically geographically diffuse and have a low attack rate. Moreover, the exposure can be subtle, hard to remember, and even unrecognized by the consumer. For example, green garnishes such as scallions or parsley may be present, but unnoticed, in a food. In addition, fresh produce has a rapidly changing geographic origin, a short-shelf life, and a rapid turnover, so implicated-products or even products originating from the same location are rarely still available by the time an outbreak has been identified. Trace-back of produce to its origins is particularly difficult because of the complex network of growers, packers, shippers, repackers, distributors, brokers, retailers, and consumers, which often involve several states and/or countries. In this complex network, produce from different suppliers can be packed together and the origin of the commodity is often lost.

Investigations of foodborne disease outbreaks involve three important components. First, the epidemiologic component identifies an association between a risk factor, e.g.: consumption of a specific food, and becoming ill. In this way, this component is said to 'point-the-way' in the investigation. The epidemiologic component also describes the outbreak in terms of who became ill, where cases occurred, the duration of the outbreak, and the clinical characteristics of the disease. Second, the environmental investigation identifies the circumstances that led to contamination, survival and growth of a microbiological agent or contamination only for chemical or physical agents of foodborne disease. The environmental investigation may also involve tracing a food back from final service through intermediary steps to initial produce in order to identify where all the relevant factors occurred. Third, the laboratory analysis is useful to support the findings of the epidemiologic and environmental investigations. Analysis of patient specimens confirms the diagnosis and analysis of food and environmental specimens substantiates the findings of the environmental investigations. Subtyping of clinical and food isolates in the laboratory provides powerful evidence of their relationship. Investigations of foodborne disease outbreaks often are unable to provide complete information for all three investigation components. In these cases investigators use expert interpretation to 'fill in the blanks' but their conclusions are less certain. The brief descriptions of produce-associated outbreaks that follow provide examples of the range of conditions under which illness has been observed.

3. Outbreaks associated with produce

3.1. Produce-associated Salmonella outbreaks

In 1955, 1979, and 1991, outbreaks of *Salmonella* infections were traced to precut watermelons (Blostein, 1993; CDC, 1979; Gayler et al., 1955). In 1990, cantaloupe from multiple sources in Mexico and Central America were linked to 295 cases of *Salmonella* Chester infections in 30 states (Ries et al., 1990). In 1991, more than 400 cases of *Salmonella* Poona infections were linked to pre-sliced cantaloupe that originated in Texas and/or Mexico (CDC, 1991b). An FDA survey conducted in 1990/1991 revealed that 35 of 3660 (0.9%) melons imported from Mexico had *Salmonella* on their surface (Madden, 1992). In 1991, the FDA recommended that food retailers wash all melons before cutting, remove the rind, maintain the temperature of the cut melons at $<7°$ C after cutting, and limit retail display to no more than 4 h.

In 1992, the produce industry implemented a voluntary Melon Quality Program which includes recommendations that hyperchlorinated water be used in all processing steps and only ice made from chlorinated water be used for transport. However, studies have shown that chlorinated water will reduce but not eliminate *Salmonella* once it is on the rind (Golden et al., 1993). Thus, chlorine is only a risk reduction factor, and other preventive measures may be needed to reduce the risk further.

In 1990, an outbreak of *Salmonella* Javiana infections involving 176 cases in Illinois, Michigan, Minnesota, and Wisconsin, was epidemiologically linked to consumption of fresh tomatoes (Wood et al., 1991). In 1993, 100 outbreak-associated cases of *Salmonella* Montevideo infections were identified in Illinois, Michigan, Minnesota, and Wisconsin, and tomatoes were again implicated as the likely vehicle (CDC, 1993). Tomatoes from both outbreaks were traced back to a packer in South Carolina; a water-bath used by the packer appeared to be a likely source of contamination of the tomatoes and the most practical point for control.

These produce-associated outbreaks represent part of a growing trend of large geographically dispersed foodborne outbreaks caused by sporadic or low-level contamination of widely distributed food items. Had the outbreaks been caused by common serotypes of *Salmonella* or had public health laboratories not been able to serotype clinical isolates, these outbreaks would not have been identified. Such episodes illustrate the complexity of fresh produce-associated foodborne outbreaks and the importance of *Salmonella* serotyping for surveillance.

Follow-up studies have shown that *Salmonella* spp. survive and grow on the surface of mature, intact tomatoes held at ambient temperature, and that growth is

rapid (reaching 10^8/gram within 24 h) in chopped ripe tomatoes at ambient temperature (Zhuang et al., 1995). Many of the patients in these outbreaks, mostly young adults, had washed the tomatoes and had removed the stem core. Did the pathogen come from inside the tomatoes? It has been shown that *Salmonella* spp. on the surface of tomatoes will be inoculated into the tomato meat during slicing, and will subsequently grow (Lin and Wei, 1997). Experiments also have shown that *Salmonella* spp. in a water bath are rapidly taken up internally through the stem scar if the water is colder than the tomatoes. The use of hyperchlorinated water will reduce but not eliminate salmonella on the external surface, and has less effect on internal *Salmonella* spp. (Zhuang et al., 1995). This has led to the recommendation that rinse water for tomatoes be hyperchlorinated and 10°F warmer than the tomatoes.

In 1995, an outbreak of *Salmonella* Hartford, *S.* Gaminara and *S.* Rubislaw infections occurred among 62 unrelated travelers at a theme park in Orlando, Florida (CDC, 1995c). Epidemiologic investigations revealed that illness was associated with drinking unpasteurized orange juice from a single manufacturer.

Salmonella Gaminara and *S.* Rubislaw were cultured from samples of unopened juice containers, but not from oranges. *Salmonella* Hartford was isolated from a toad. Amphibians were in close proximity to the processing building and the facility was excessively open to the environment. The isolation of these serovars from patients, product, and the processing environment established the cause of the outbreak. Additional microbiological analyses of samples obtained before and after a thorough cleaning and sanitizing of the processing facility provided evidence of inadequate sanitation practices (Parish, 1997). Following the outbreak, the theme park began using only pasteurized orange juice.

A 1995 outbreak of *Salmonella* Stanley infections involving 23 US states and Finland was epidemiologically linked to consumption of alfalfa sprouts (Mahon et al., 1997). The outbreak strain was distinguished by its unique antibiotic resistance pattern and unique molecular characteristics. The implicated sprouts were traced through nine growers and one distributer to one Dutch shipper. The Finnish sprouts were also traced to seeds from the same Dutch shipper. Thus, it appears that an international outbreak was the result of contaminated seeds distributed to two continents.

In 1996, an outbreak of *Salmonella* Newport infections associated with sprouts in Oregon and British Colombia was traced to seed from the same Dutch shipper (Van Beneden, 1996). Follow-up studies have shown that while populations of *S.* Stanley could be greatly reduced, elimination of this organism from alfalfa seeds may not be reliably achieved with traditional disinfection procedures. If *S.* Stanley is present on seeds at the initiation of the sprout production process, populations exceeding 10^7 CFU/g can develop and survive on mature sprouts exposed to handling practices used in commercial production and marketing. Potential solutions to this problem include avoiding contamination during seed production and distribution and using a 2–4 mg/ml chlorine soak treatment of alfalfa seeds before germination. The chlorine soak treatment greatly reduces populations of *S.* Stanley, and possibly other salmonellae, while not adversely affecting germination (Jaquette et al., 1996).

The CDC has detailed two further outbreaks of salmonellosis attributed to the consumption of alfalfa sprouts, one in 1996 and one in 1997 (letter from Griffin, Slutsker and Tauxe to Fred Shank, dated July 29, 1997). In 1996, more than 500 cases of *Salmonella* Montevideo and >100 cases of *Salmonella* Meleagridis infection occurred in California, which were linked to the consumption of alfalfa sprouts from a single sprouter. Cultures of sprouts from the facility yielded *S.* Meleagridis. A second outbreak attributed to *Salmonella* Infantis and *Salmonella* Anatum occurred between February and June 1997, in Kansas and Missouri. Approximately 80 cases of *S.* Infantis and 10 cases of *S.* Anatum were reported. Cultures of seeds and environmental swabs from the implicated sprouter yielded both *S.* Infantis and *S.* Anatum.

3.2. Shigella flexneri from scallions

In 1994, a multi-state outbreak of shigellosis was epidemiologically linked to consumption of scallions (Cook et al., 1995). The outbreak was detected because of a seven-fold increase in the numbers of *Shigella flexneri* 6A cases reported in Illinois. Sixteen culture-confirmed cases of *Shigella* infection were traced to green onions from five US and at least one Mexican farm. Barriers to prevent the outgrowth of pathogens were not used, though several potential sources of contamination of green onions during growth, harvest, and shipping were found. Because *S. flexneri* 6A is very rare in the US and relatively common in Mexico, contamination at the time of harvest or during packaging in Mexico is a plausible point of contamination in this outbreak. This outbreak illustrates how the occurrence of unusual and distinctive pathogens can result in the detection of an outbreak.

3.3. Produce-associated outbreaks of E. coli O157:H7 infections

In the US, *E. coli* O157:H7 infections are usually associated with ground beef. However, other foods and drinking water have also been implicated in outbreaks of infections caused by this pathogen. In 1991, an outbreak of *E. coli* O157:H7 infections (23 cases) in Massachusetts was associated with consumption of apple

cider prepared from a cider mill (Besser et al., 1993). Ninety percent of the apples used in the cider were 'drops' – apples collected from the ground. Contamination may have occurred prior to harvest or at harvest. Low pH and temperature have been presumed to be barriers to survival and growth of *E. coli* O157:H7; however, prolonged survival has been documented at a pH less than 4.0 and at refrigerator temperature (8° C) in experimentally contaminated apple cider (Zhao et al., 1993). To prevent future similar outbreaks, the practice of using 'drops' in cider and using unprocessed manure and manure composted under the current state of composting practices as fertilizer should be reviewed. Also, raw produce should be effectively cleaned before being processed for human food and should be stored under conditions that will protect against contamination and minimize deterioration. Additional barriers may be achieved by pasteurization or by including permissible antimicrobials.

In 1995, an outbreak of *E. coli* O157:H7 infections involving at least 29 people in Montana was epidemiologically linked to consumption of leaf lettuce (CDC, 1995a). The mechanism of contamination was not determined. However, the lettuce was irrigated with surface water, and investigations of area grocery stores revealed unsanitary leaf lettuce handling methods. Also in 1995, an outbreak of *E. coli* O157:H7 infections involving 30 persons attending a Boy Scouts gathering in Maine was epidemiologically linked to iceberg lettuce (CDC, 1995b). It is not known where or when the lettuce was contaminated. However, it is possible that cross-contamination from meat products may have occurred during food preparation or storage.

Two additional *E. coli* O157:H7 outbreaks involving mesclun mix lettuce were reported to the CDC in 1996, one involving at least 26 patients in Illinois and the other involving 20 patients in Connecticut. Isolates from both outbreaks were indistinguishable by pulsed-field gel electrophoresis (PFGE) molecular analyses, indicating that the two outbreaks might have the same origin. Epidemiologic data from both outbreaks implicated leafy salad vegetables that may have originated from the same grower. Although *E. coli* O157:H7 was not isolated from the suspected farm, numerous good manufacturing practice (GMP) violations were observed in the on-farm processing facility and potential sources of bovine and avian fecal contamination were present (California Department of Health Services, Food and Drug Branch).

In October and November 1996, an outbreak of *E. coli* O157:H7 infections associated with consumption of a commercial brand of unpasteurized apple juice occurred in western North America (CDC, 1996c). Initially a cluster of *E. coli* O157:H7 infection, mainly in young children, was noted in King County, Washington. Investigations determined that these cases were epidemiologically linked to the consumption of fresh apple juice. The apple juice was unpasteurized and was intended for consumption within two weeks of processing. The product was distributed predominantly in the western US and British Columbia. A total of 70 cases of apple juice-associated *E. coli* O157:H7 infections were identified among residents of California, Colorado, Washington, and British Columbia. More than half of the cases involved patients aged five years or younger. Fourteen patients developed hemolytic uremic syndrome (HUS), and one child died. An unopened container of epidemiologically-implicated unpasteurized apple juice yielded *E. coli* O157:H7 with the same molecular subtype as the patient isolates. Comparison of the DNA'fingerprints' of the juice isolate with the patient isolates by restriction enzyme digest showed they were indistinguishable. The manufacturer's handling processes included inspection and hand sorting of all incoming fruit, and washing (and probably brushing) of apples prior to pressing. Dropped apples were not considered acceptable for juice production. An environmental investigation of the facility producing the apple juice did not identify the source of contamination and *E. coli* O157:H7 was not isolated from any of the numerous cultures taken from the facility, its water sources, or incoming apples.

This outbreak demonstrated what had been suspected from the earlier apple cider-related outbreaks in New England; that *E. coli* O157:H7 can survive in juice at acidic pH values and at refrigeration temperature. Apparent lack of growth is an insufficient indicator of the presence of bacterial pathogens because of the short shelf-life (two to three weeks) of these fresh juice products and the very low infectious dose of *E. coli* O157:H7. Because unpasteurized juice does not have a terminal pathogen kill step in its production and traditional preservatives such as 0.1% sorbate have been ineffective in killing *E. coli* O157:H7, fresh juices, particularly apple juice, must be regarded as beverages potentially at risk for microbial contamination and disease transmission. The FDA is investigating whether methods other than pasteurization are capable of preventing fresh juices from transmitting microbial pathogens.

During the summer of 1996, the largest known outbreaks of *E. coli*, O157:H7 to date occurred in Japan. More than 6000 cases were reported, mainly in central and southern Japan. Molecular analysis showed that this was not a single clone, but represented multiple PFGE subtypes. The largest outbreak affected over 4000 school children in and around Sakai City where there were four deaths. Infection appeared to have been transmitted through the consumption of raw radish sprouts served on either of two days in school lunches. These lunches were prepared in central kitchens with a subsequent peripheral distribution to the schools in high volume. The contaminated radish sprouts unfortunately

then gained access to an extremely large population of school children. Although considerable environmental and food investigation yielded at least three *E. coli* O157:H7 isolates, none matched the PFGE pattern of the isolates from the Sakai City school children. No radish sprouts were available from the implicated dates on which they were served, and the source of the contamination remains unknown (Michino et al., 1996; Michino, 1997).

More recently, outbreaks of alfalfa sprout-associated infections with *E. coli* O157:H7 have been identified in the US (CDC, 1997g). Outbreaks of *E. coli* O157:H7 occurred between the months of June and July 1997 in two states. A total of 60 cases were reported in Michigan, all traced to a single Michigan alfalfa srouter. Virginia reported 48 cases also traced to a single Virginia sprouter. Both sprouters utilized alfalfa seeds from the same distributor lot, and PFGE patterns from patients involved in both outbreaks were indistinguishable. Microbiological examination of seeds and aseptically sprouted seeds have failed to isolate this organism from either source.

3.4. Enterotoxigenic E. coli (ETEC) from carrots

Two clusters of ETEC infections occurred in 1993 in Rhode Island (an airline flight with 47 cases) and New Hampshire (a lodge with 121 cases) (CDC, 1994). ETEC serotype O6:non-motile (NM) that produced ST and LT toxins was isolated from several cases in both groups. Plasmid profiles of these strains were identical to each other but differed from those of 10 other ETEC O6:NM strains from other sources. Epidemiologic investigation of patients infected with strains that were indistinguishable on the basis of serotype and plasmid profile implicated carrots as the source of infection. This outbreak is an example of the value of molecular subtyping in epidemiologic investigations.

3.5. Botulism from chopped garlic

A 1985 epidemiologic investigation of a multinational outbreak, constituting the second largest outbreak of botulism reported in North America, implicated an unexpected vehicle for botulism intoxication (St Louis et al., 1988). All 36 identified cases of botulism type B had eaten at a single restaurant in Vancouver, British Colombia, and the implicated food item was garlic butter made from commercially bottled chopped garlic in soybean oil. The chopped garlic was a sundried, rehydrated aqueous mixture with a pH above 4.6 without chemical or acidifying additives, and was labeled with instructions to refrigerate. However, it had been stored at room temperature conditions conducive to the outgrowth of *Clostridium botulinum* spores. The production of botulinum toxin in bottles of the commercial garlic at 25°C after experimental inoculation with *C. botulinum* spores showed that it must be refrigerated to prevent botulism. After three more cases of botulism were linked to chopped garlic in oil in New York in 1989, the FDA issued the recommendation that garlic-in-oil products should contain acidifying agents such as phosphoric or citric acid to add additional obstacles to *C. botulinum* growth. Non-preserved products should be kept refrigerated at all times from manufacturing to consumption to prevent botulism.

3.6. Cryptosporidium from apple cider

A 1994 outbreak of cryptosporidiosis in Maine involving 160 primary cases and 53 secondary cases was associated with drinking hand-pressed apple cider (Millard et al., 1994). Oocysts of *Cryptosporidium parvum*, a zoonotic protozoan parasite, were detected in the stools of 50 to 56 patients examined, in the apple cider, on the cider press, and in a fecal sample from a calf on the farm that supplied the apples. A similar outbreak of apple cider-associated infections with *Cryptosporidium* occurred in New York in October 1996 (CDC, 1997a). In this outbreak, the strength of the epidemiologic association between drinking cider from the implicated cider mill and illness was very strong and accounted for infections in 17 of 18 case households enrolled in the case–control study. However, *Cryptosporidium* was not identified in environmental or product samples, and previously identified risk factors of using drops and manuring the orchard, were reported not to have been practiced.

3.7. Cyclospora associated with raspberries

Cyclospora cayetanensis is a newly recognized and incompletely understood protozoan parasite. *Cyclospora cayetanensis* is known to be spread by indirect fecal–oral transmission, but oocysts are reportedly not immediately infectious, so direct person-to-person spread is unlikely. The first recognized outbreaks of infections with this emerging pathogen were initially attributed to drinking water. Related coccidia are host specific, and this pathogen is believed to infect only humans; however, the possibility of an animal reservoir for *C. cayetanensis* is being explored.

In the summer of 1996, a large epidemic of *C. cayetanensis* infections occurred in the US and Canada (Herwaldt and Ackers, 1997). A total of more than 1,450 cases of diarrheal disease were reported in 20 States, Washington DC, and two Canadian provinces; many of these infections were laboratory confirmed despite the diagnostic difficulties that exist for this pathogen. Several epidemiologic investigations were conducted, and a strong association was demonstrated between consumption of raspberries from Guatemala and *Cyclos-*

pora infection in a large number of defined case clusters. While the growing areas of some of the implicated raspberries were identified, no *Cyclospora* was detected in specimens from these sites. Investigations are hampered by the limited methods available for detecting *Cyclospora* from food or water (which are of unknown sensitivity), unavailability of berries from implicated shipments, and unknown risk factors during production, picking, and packaging. In the summer of 1997, raspberries, mesclun lettuce, and basil/basil-containing products were implicated in numerous clusters of infections with *C. cayetanensis* (CDC, 1997c; CDC, 1997d; CDC, 1997e; CDC, 1997f). The implication of three different fresh produce vehicles of infection highlights the importance of extending our knowledge and understanding of *Cyclospora* and strengthening prevention and control measures to ensure the safety of fresh produce.

3.8. Perspectives gained from outbreak investigations

An important function of outbreak investigations is to determine the point of contamination so that future outbreaks can be prevented. Of the 27 outbreaks described above, the investigations clearly identified a point of contamination in two (Table 2). In many of the other outbreaks, the investigations identified multiple links in the chain from growing to preparation in which contamination may have occurred, but a single point of contamination could not be determined. In the case of cider-associated cryptosporidiosis, finding *C. parvum* in cattle on the farm that produced the apples and in leftover cider strongly suggests that the agent was introduced during growing and harvesting of the apples; however, microbiologic confirmation was not available to lend further support to this conclusion.

From 1990 to 1996, 122 confirmed foodborne outbreaks were reported in Minnesota. Fresh produce items were the most frequently implicated food vehicle, and accounted for 37 (30%) outbreaks (Appendix A, Table 7). In eight (22%) outbreaks the implicated fresh produce items were served on salad bars. The most common agent associated with fresh produce outbreaks was Norwalk-like virus, which caused 20 (54%) outbreaks (Appendix A, Table 8). The source of contamination for most of these outbreaks was a sick person who had handled the implicated produce item at the point of preparation. In several cases, asymptomatic foodhandlers with ill family members were the most likely source of contamination. *Salmonella* spp. accounted for six (16%) confirmed produce-associated outbreaks, and four (50%) of the salad bar-associated outbreaks.

Another finding from outbreak investigations has been to note that in several instances, agricultural

Table 2
Examples of produce-associated outbreaks by identified point of contamination.

Stage of food chain	Produce item	Pathogen	Reference
Growing and harvesting	Unpasteurized apple cider	*E. coli* O157:H7	Besser et al. 1993
	Chopped garlic	*C. botulinum*	St Louis et al. 1988
Undetermined and/or multiple	Sliced watermelon	*Salmonella* spp.	Gayler et al. 1955
	Sliced watermelon	*Salmonella* spp.	CDC, 1979
	Sliced watermelon	*Salmonella* spp.	Blostein, 1993
	Sliced cantaloupe	*Salmonella* spp.	Ries et al. 1990
	Sliced cantaloupe	*Salmonella* spp.	CDC, 1991b
	Tomatoes	*Salmonella* spp.	Wood et al. 1991
	Tomatoes	*Salmonella* spp.	CDC, 1993
	Unpasteurized orange juice	*Salmonella* spp.	CDC, 1995c
	Alfalfa sprouts	*Salmonella* spp.	Mahon et al. 1997
	Alfalfa sprouts	*Salmonella* spp.	Van Beneden, 1996
	Green onions	*Shigella* spp.	Cook et al. 1995
	Alfalfa sprouts	*E. coli* O157:H7	CDC, 1997g
	Leaf lettuce	*E. coli* O157:H7	CDC, 1995a; CDC, 1995b
	Unpasteurized apple juice	*E. coli* O157:H7	CDC, 1996c
	Unpasteurized apple cider	*E. coli* O157:H7	CDC, 1997a
	Radish sprouts	*E. coli* O157:H7	Michino, 1996
	Carrots	Enterotoxigenic *E. coli*	CDC, 1994
	Frozen strawberries	Hepatitis A virus	Niu et al. 1992
	Frozen strawberries	Hepatitis A virus	CDC, 1997b
	Unpasteurized apple cider	*C. parvum*	Millard et al. 1994
	Unpasteurized apple cider	*C. parvum*	CDC, 1997a
	Raspberries	*C. cayetanensis*	CDC, 1996a,b
	Raspberries	*C. cayetanensis*	CDC, 1997d,e
	Mesculn lettuce	*C. cayetanensis*	CDC, 1997e
	Basil/basil-containing products	*C. cayetanensis*	CDC, 1997f

Table 3
Examples of produce-associated outbreaks with probable points of contamination.

Stage of food chain	Product	Pathogen	Reference
Growing and harvesting or Handling and processing	Cantaloupes	*Salmonella*	Ries et al. 1990; CDC, 1991b
Handling and processing	Tomatoes	*Salmonella*	Wood et al. 1991; CDC, 1993
Growing and harvesting or Handling and processing	Scallions	*Shigella*	Cook et al. 1995
Growing and harvesting	Sprouts	*Salmonella*	Mahon et al. 1997
Growing and harvesting or Handling and processing	Apple cider	*Cryptosporidium parvum*	Milard et al. 1994; CDC, 1997a
Processing	Lettuce	*Shigella sonnei*	Davis et al. 1988

workers were the likely source of the pathogen contaminating the fresh produce (Ackers et al., 1997; Niu et al., 1992; Reid and Robinson, 1987; Rosenblum et al., 1990). Fresh produce can be particularly vulnerable to this means of contamination when, as for some types of produce, there are virtually no critical control points for pathogen elimination between harvesting and consumption. For this reason, persons who harvest and/or process fresh produce should perhaps be viewed as food handlers rather than agricultural workers.

These outbreaks clearly indicate that all stages from primary production to preparation are important points of introduction, and that we must look for new pathogens when investigating fresh produce-associated diseases and examine the role of produce when we investigate outbreaks caused by new pathogens. These outbreaks also demonstrate the need for improved diagnostic methods for foodborne protozoa, and the importance of reacting appropriately to strong epidemiologic data, even without laboratory confirmation, to interrupt the course of an epidemic.

In some investigations, plausible possibilities for the source of contamination can be determined. Table 3 gives examples of nine produce-associated outbreaks in which probable points of contamination were identified.

4. Microbial ecology of foodborne pathogens associated with fresh fruits and vegetables

4.1. Introduction

As indicated by the epidemiologic investigations discussed above, diseases associated with fresh fruits and vegetables are primarily those transmitted by a fecal–oral route. This implies that control of fecal contamination is a primary concern, and much of the following discussion focuses on its control. However, it should be noted that not all pathogens can be correlated with indicators of fecal contamination, and there are other pathogens whose primary source is not feces (Nguyen-the and Carlin, 1994; Monge and Chinchilla, 1996). While fresh produce can serve as the source of all classes of foodborne pathogens (bacteria, viruses, protozoa, fungi, and helminths), the greatest concerns are generally associated with pathogenic bacteria; the risk of disease is amplified because of potential growth prior to consumption. The growth, survival, and inactivation of microorganisms on fresh fruits and vegetables is dependent on the interaction of four broad factors: (1) the characteristics and capabilities of the microorganisms present; (2) the physiological state of the plant tissue and its inherent resistance to microbial metabolic processes; (3) the characteristics of the environment surrounding the plant tissue (e.g., pH, water activity, and atmosphere composition); and (4) the effect of food processes and practices on microbial populations or plant metabolism.

It is important to note that the factors discussed below can reduce the risks associated with produce but cannot eliminate them. The foods in question are raw agricultural commodities that do not receive any 'lethal' treatment specifically designed to kill all pathogens prior to consumption. It must be assumed that even under the best production, processing, distribution, and marketing conditions, a small percentage of produce will harbor one or more pathogens. This implies that the focus of activities must be realistically directed toward risk reduction and not elimination. This also implies that care must be exercised in appropriately communicating those risks to consumers, and that customer education is a component of an overall control strategy.

The lack of a lethal treatment at any step between farm production and consumption also means that pathogens introduced at any point may be present when the produce is consumed. For most enteric pathogens, the ultimate source will be the feces of an animal or human. However, the specific means through which produce is contaminated are varied, and include both direct and indirect transmission via animals, soil, water, and human handling.

The nature of microbiological contamination of fresh produce also implies that the potential for microbiological testing to identify and permit the removal of contaminated product is severely limited. Analysis of produce for specific pathogens or indicators of fecal contamination may detect high frequency occurrence of gross contamination. However, the sporadic nature of most contamination will make it unlikely that microbiological testing will, with any degree of assurance, effectively identify contaminated products. This is

particularly true when the level of pathogen control is increased, resulting in a decrease in the frequency and extent of pathogen isolations. However, it should be noted that this does not preclude the use of appropriate microbiological testing to assess the effectiveness of control programs. Again, these are critical concepts that must be effectively communicated to consumers and producers.

4.2. Production sources

While the frequency of isolation may be changed as a result of subsequent processing, to a great extent the microflora of market fruits and vegetables reflect the species present at the time of harvest (Nguyen-the and Carlin, 1994). This presumably holds true for a significant portion of pathogen isolations associated with fruits and vegetables. The microflora of fresh fruits and vegetables is diverse, but is predominately Gram-negative bacteria. The levels of bacteria on plants in the field can vary widely, and even on a single plant the levels of bacteria on individual leaves can differ substantially. Microbial contamination of plant tissue is largely associated with the surfaces of fruits or vegetables, and the inner tissue of sound vegetables and fruit are often considered sterile. For this reason, leafy vegetables which have the greatest surface area are often the most heavily contaminated. However, numerous investigators have reported isolating low levels of bacteria from internal tissues of apparently intact vegetables (Lund, 1992; Robbs et al., 1996). It has been demonstrated that the application of bacteria to the surface of fruits will result in their internalization over time (Samish and Etinger-Tulczynska, 1963).

Anything in the production environment that comes in contact with the plant has the potential to be a pathogen source. Irrigation water, animals, farm workers, and the soil are particularly important. Soil that has not been contaminated with feces is generally not a primary source of enteric microorganisms. However, *C. botulinum*, *Clostridium perfringens*, and *B. cereus* can be isolated from soils free of fecal contamination, and may be found commonly associated with fresh vegetables (Hauschild, 1989; Labbe, 1989; Roberts et al., 1982). Likewise, *Listeria monocytogenes* is considered environmentally ubiquitous, and can be found associated with decaying vegetation and soil (Welshimer, 1968; Welshimer and Donker-Voet, 1971; Fenlon, 1985). Coliforms can be found in association with both soil and decaying vegetation, but these are largely restricted to coliforms of non-fecal origin such as *Enterobacter* spp. and *Klebsiella* spp. Significant levels of *Escherichia coli* and related enteric microoganisms of fecal origin are thought to be limited to soils that have been fecally contaminated (Geldreich et al., 1962), and are transitory in nature. The normal association of thermotolerant coliforms such as *Klebsiella* with produce limits the value of 'fecal coliforms' as an indicator of fecal contamination. Indicators more specifically associated with fecal contamination (e.g., *E. coli*) are more appropriate. The survival of enteric bacteria in the soil is dependent on inoculum, soil type, moisture retention, pH, microbial antagonisms and nutrient availability (Geldreich and Bordner, 1971). It has generally been concluded that except for the soilborne pathogenic bacteria mentioned above, soil *per se* is not an important source of human pathogens on plants. However, the presence of fecally transmitted pathogens on fresh produce is likely to occur if improperly composted sewage or manure is used as a fertilizer.

One important source of fecal contamination appears to be related to the quality of water used for irrigation. It has long been known that the use of irrigation water with high levels of enteric bacteria, viruses, protozoa, or helminths results in increased frequency of pathogen isolations from harvested produce (Norman and Kabler, 1953; Dunlop and Wang, 1961). The extent of contamination is dependent on both the type of irrigation and produce. The greatest contamination is generally associated with leafy vegetables which provide large surface areas and topographical features that foster the attachment or entrapment of microorganisms. High relative humidity favors the survival and spread of bacteria on plant surfaces (Leben, 1988). Bacteria can adhere quite tenaciously and cannot be easily removed by gentle washing (Preece and Wong, 1981). Irrigation techniques that subject the plant to direct contact with contaminated water increase the risk of contamination. Accordingly, spray irrigation would be expected to increase contamination in comparison to either drip irrigation or flooding. Irrigation water can become polluted either through the direct introduction of sewage, or through non-point pollution sources such as ground water runoff. For example, drainage and run-off from animal pens after rain can lead to the localized contamination of irrigation sources. Identification of point and non-point sources of microbial contaminants is the foundation for the development of management practices for reducing microbiological risks associated with irrigation waters (CDC, 1997h). The risk of fecal contamination can be reduced by treatment of contaminated water sources prior to use for irrigation (Robinson and Adams, 1978).

Wild and domestic animals, including mammals, birds, reptiles, and insects, are another source of pathogenic bacteria in agricultural environments. Salmonellae have been isolated from the intestinal tracts of most warm-blooded and many cold-blooded animals. Birds can be a particularly important contamination source because of their ability to transmit bacteria over substantial distances. For example, it has been reported that gulls feeding at sewage works are an important vector for subsequently introducing *L. monocytogenes* and enteric bacteria into the agricultural environment

(Fenlon, 1985). Insects are another potential vector for transfer of fecal material from sites of sewage or animal manure accumulation to produce production areas (Geldreich et al., 1964). There is some indication that pollinating insects can serve as a means of transmitting enteric bacteria to flowers. Once contaminated, developing fruit may internalize the bacteria eliminating their accessibility to surface decontamination steps. A significant portion of the resident populations of rodents, rabbits, and other commonly occurring mammals in agricultural production areas are likely to harbor enteric pathogens and are potential sources of fecal contamination, either through direct contamination of the field or via contamination of irrigation waters (Geldreich and Bordner, 1971). Allowing domestic animals access to orchards can result in the contamination of fresh fruits and treenuts, particularly if these are gathered after having fallen to the ground. For example, such practices may be linked to the presence of *Salmonella* and *E. coli* O157:H7 in apple cider and apple juice (CDC, 1975; Goverd et al., 1979; Besser et al., 1993).

As with animals, it must be assumed that a portion of the humans in the farm environment harbor one or more enteric pathogens and thus may play a role in the contamination of fresh produce. The potential for transferring fecal contamination to the surface of produce is increased when there is a lack of suitable sanitary hand-washing facilities in the production area. This appears to be particularly important in the transmission of enteric viruses such as hepatitis A, wherein growth of the pathogen on the produce is not important. Human involvement on the farm is particularly important during harvest and subsequent handling of the fruits and vegetables.

4.3. Processing of fresh fruits and vegetables

Most fruits and vegetables receive some degree of processing before being placed in commercial distribution. Increasingly, this involves some degree of minimal processing to increase the convenience and value of the product. Some of the key microbiological characteristics of fresh produce processing are: (1) there is an increased presence of cut surfaces or damaged plant tissues which supply nutrients for microbial growth; (2) the degree of processing is insufficient to ensure sterility or even microbiological stability; (3) the plant tissue remains metabolically active; and (4) there is a mixing and confinement of the product (Nguyen-the and Carlin, 1994). Even in the absence of cutting, the initial quality and subsequent handling of fresh fruits and vegetables appears to strongly influence its microbiological safety. For example, Wells and Butterfield (1996) reported that the rate of *Salmonella* isolation from produce affected by bacterial soft-rot was approximately double that of intact fruits and vegetables. Further, they demonstrated that salmonellae grew more quickly on potato, carrot, and green pepper when co-cultured with *Erwinia carotovora* or *Pseudomonas viridiflava*, two major causes of bacterial soft-rot.

Bacteria, protozoa, and viruses of public health concern can survive for extended periods on fresh produce, and under favorable conditions specific fresh fruits and vegetables may support the growth of pathogenic bacteria (Table 4). Concerns about bacteria pathogenic to humans and associated with fresh produce are amplified when the fruit or vegetables support microbial growth. The two principal determinants of growth of pathogens on produce are pH and storage temperature.

Table 4
Examples of survival and growth of pathogenic microorganisms on/in fresh produce.

Pathogen	Produce/conditions	Reference
Survival		
E. coli O157:H7	Apple cider 8°C	Zhao et al. 1993
Shigella spp.	Lettuce 5°C/3 d	Davis et al. 1988; Satchell et al., 1990
Salmonella spp.	Melons 22–27°C/6 h	Escartin et al. 1989
	Sprouts 5°C/10 d	Jaquette et al. 1996
Campylobacter jejuni	Sliced melon 25–29°C/6 h	Castillo and Escartin, 1994
Enterovirus	Fresh produce 4°C/25–30 d, 22°C/5–25 d	Bagdasargan, 1964; Badawy et al., 1985
Growth		
Shigella spp.	Lettuce 22°C	Davis et al. 1988
	Melon 22–26°C	Escartin et al. 1989
	Cabbage 22°C	Satchell et al. 1990
Salmonella spp.	Tomatoes 20°C	Zhuang et al. 1995
	Melons 23°C	Escartin et al. 1989; Golden et al. 1993
	Sprouts 21°C	Jaquette et al. 1996
E. coli O157:H7	Melons 25°C	Del Rosario and Beuchat, 1995
	Shredded lettuce 12°C	Abdul-Raouf, 1993
	Sliced cucumbers 21°C	Abdul-Raouf et al. 1993
Yersinia enterocolitica	Fresh produce 4°C	Darbas et al., 1985
Listeria monocytogenes	Fresh produce 4°C	Beuchat and Bracket, 1990; Berrang et al. 1989

In addition, some plant tissues have naturally occurring antimicrobials that provide varying levels of protection against the growth of pathogens (Lund, 1992).

Most vegetables have a pH of 4.5 or higher and as such are able to support the growth of a variety of bacteria. In such products, storage temperature then becomes the principal factor that controls microbial growth. Many fruits (e.g., apples, oranges) are more acidic than vegetables and do not support the growth of human pathogens. However, pH can still be important since bacteria are inactivated more quickly at lower pH values. This can be particularly important for products that rely on their low pH to inactivate highly infectious enteric pathogens such as *Shigella* or *E. coli* O157:H7 (Miller and Kaspar, 1994). A number of melons and soft fruits have pH values that are substantially higher than 5.0 and will support the growth of a variety of pathogenic bacteria (Gayler et al., 1955; Escartin et al., 1989; Nguyen-the and Carlin, 1994; Del Rosario and Beuchat, 1995; Beuchat, 1996). For example, honeydew melon, cantaloupe, and watermelon have pH values ranging from 6.3 to 6.7, 6.2 to 6.5, and 5.2 to 5.6, respectively (Lund, 1992). Tomatoes are of particular interest because of their extensive use, handling practices, and the general misconception that they do not support pathogen growth. The pH of tomatoes is dependent on both the variety and state of ripeness; the pH range for tomatoes is 3.4–4.8 (Lund, 1992). Recently, it has been demonstrated that *Salmonella* can grow on sliced or cut tomatoes at pH values as low as 3.99 (Asplund and Nurmi, 1991; Wei et al., 1995; Zhuang et al., 1995).

For produce whose pH permits the growth of pathogenic bacteria, the product's relative safety will be determined by storage temperature. In general, the minimum temperature for the growth of most mesophilic enteric pathogens is 8–10°C, though this may be higher when pH values approach the minimum values that support growth. Often, unrefrigerated products will spoil before there has been extensive growth of pathogens; however, this cannot be relied on as an effective control (Piagentini et al., 1997). While adequate refrigeration is an important safeguard, it is not absolute because a number of psychrotrophic pathogens such as *L. monocytogenes*, *Y. enterocolitica*, and *Aeromonas hydrophila*, can grow in fresh vegetables. In particular, *L. monocytogenes* has been shown to grow on a variety of vegetables, with growth at refrigeration temperatures being dependent on the interaction between pH and temperature (Steinbruegge et al., 1988; Berrang et al., 1989; Beuchat and Brackett, 1990; Beuchat and Brackett, 1991; Kallander et al., 1991; Carlin and Nguyen-the, 1994; García-Gimeno et al., 1996).

After harvesting, fresh produce has a diverse miocroflora and can be heavily contaminated. If there is a significant delay in transport to the processing facility, there can be substantial bacterial replication when temperatures are elevated, and humid conditions are maintained (Spittstoesser, 1970). In general, effective processing will reduce the overall microbial load associated with produce; however, if care is not taken, it can lead to the dissemination of pathogens via cross-contamination (Velani and Roberts, 1991). Since most microbial contamination is on the exterior surfaces of fruits and vegetables, surface treatments can reduce the overall degree of contamination. For example, removal of the exterior leaves from heads of cabbage substantially reduces the overall levels of bacteria (Keipper and Fred, 1930). Washing will remove a portion of the bacteria from the surface of produce but cannot assure complete removal of pathogens (Garg et al., 1990; Beuchat, 1996). Washing can also help remove cell contents released during slicing or shredding that help support the growth of microorganisms (Bolin et al., 1977). Care must be taken to ensure that this does not actually aggravate food safety concerns. If pathogens are not inactivated or physically removed during the washing process, pathogens can be spread so that a significant portion of the produce is contaminated instead of a sporadic plant (Nguyen-the and Carlin, 1994). Chlorine is commonly used to help prevent cross-contamination and increase the efficacy of washing, but chlorine does not completely assure pathogen elimination (Beuchat and Brackett, 1991; Park and Sanders, 1992; Beuchat, 1996; Wei et al., 1995; Zhuang et al., 1995). Chlorine dioxide, hypochlorite, and trisodium phosphate have also been evaluated for their potential use for disinfecting produce, but again cannot assure pathogen elimination (Costilow et al., 1984; Park et al., 1991; Zhuang and Beuchat, 1996). For some commodities the temperature of wash water should be greater than that of the produce or the pressure differential will result in the aspiration of bacteria into the plant material (apples, celery, tomatoes) (Lund, 1992; Robbs et al., 1996). The use of edible coatings (e.g., hydroxypropyl methylcellulose) has been shown experimentally to reduce the levels of salmonellae on the surface of tomatoes (Zhuang et al., 1996).

It is important to note that certain fresh soft fruits (e.g., raspberries, strawberries) and vegetables (e.g., green peppers) cannot tolerate exposure to water. For these products, disinfective rinses are not a realistic option at the processing level, and alternative interventions are needed. Currently, this is largely limited to low-dose ionizing radiation (<1 kGy) used for shelf life extension of products such as strawberries. At these doses, there would be some reduction in the levels of pathogenic enteric bacteria and a more substantial reduction in pathogenic protozoa (e.g., *Cyclospora*, *Cryptosporidium*). Somewhat higher doses (1–3 kGy) would be needed to control bacterial pathogens such as *Salmonella* and *E. coli* O157:H7.

Major sources of in-plant contamination of minimally processed produce are shredding and slicing/cutting operations (Garg et al., 1990; Jöckel and Otto, 1990). Some of this may be an artifact, reflecting the increased accuracy of estimating the true mean of a microbial population achieved by normalizing the distribution of microorganisms on a food (Kilsby and Pugh, 1981). However, experimental work has demonstrated clearly that passing a knife through a contaminated surface inoculates the newly exposed surfaces of cut produce (Lin and Wei, 1997). Shredding and slicing equipment are often 'hard to clean' and identified as important loci for the accumulation of pathogens in processing environments (Lainé and Michard, 1988).

As in the agricultural production environment, human contact at processing facilities and during subsequent distribution and marketing can lead to contamination of fresh fruits and vegetables (Davis et al., 1988).

The shelf life of minimally processed food can be extended significantly through the use of controlled atmosphere storage or modified atmosphere packaging. This takes advantage of the fact that elevated levels of carbon dioxide retard the growth of fungi and Gram-negative psychrotrophs. This leads to the modification of microflora to slower growing Gram-positive bacteria that produce less deleterious quality changes. However, there has been concern that these technologies could retard spoilage but not have a similar effect on the growth of pathogens. This concern was originally focused on the potential growth of *C. botulinum*. A high CO_2 atmosphere was found to support the growth of *C. botulinum* type A on shredded cabbage stored at room temperature. However, it appears that the incidence of *C. botulinum* in modified atmosphere packaged vegetables is relatively low (Lilly et al., 1996). In some instances the use of plastic wraps in combination with the rapid respiratory activity of plant tissue can generate a high CO_2 atmosphere. This was the case with fresh mushrooms, where holes in the wrap were used to keep the atmosphere from becoming so anaerobic that *C. botulinum* can produce toxin (Sugiyama and Yang, 1975; Kautter et al., 1978; Solomon et al. 1990).

More recently, concern about modified atmosphere-packaged produce has focused on *L. monocytogenes*. Several groups of investigators have demonstrated that modified atmospheres can increase shelf life without affecting the growth of *L. monocytogenes*, (Berrang et al., 1989; Beuchat and Brackett, 1990; Kallander et al., 1991; García-Gimeno et al., 1996). This is of particular concern because of the pathogen's ability to grow at refrigeration temperature. The suppression of the microflora could lead to a situation where in adequately refrigerated produce, *L. monocytogenes* could reach high levels before the product becomes overtly spoiled. Elimination of background microflora through the use of chemical disinfection has also been reported to increase the growth of *L. monocytogenes* on produce (Carlin et al., 1996).

4.4. Food service

Outbreak investigation results indicate that contamination of produce by food service workers has been a significant factor in foodborne illness associated with consumption of fresh produce (data provided by Michael Osterholm, M.D.). This may be directly related to the significant amount of direct hand contact which occurs in food service and retail establishments during the preparation of fresh produce items for serving. Since the preparation of produce items at food service and retail establishments typically does not involve application of a treatment designed to inactivate microorganisms, food worker hygienic practice can be expected to have a direct influence on the microbiological characteristics of fresh produce items.

Cross-contamination in food service establishments has also been shown to contribute to outbreaks of foodborne illness associated with consumption of fresh produce (CDC, 1998). Potential for contamination through this route should be addressed by food service and retail establishments.

4.5. Fruits and vegetables in the home

How produce is held and handled within the home will greatly influence its shelf life or contribute to the possibility of foodborne illness. Unpublished survey data (Procter and Gamble, 1993;Procter and Gamble, 1997) indicate that a majority of consumers rinse at least a portion of their produce. Typically tap water is used just prior to eating or cooking. The frequency that consumers rinse products varies among commodities. More aggressive cleaning techniques are employed by a portion of consumers for certain commodities (e.g., scrubbing of potatoes).

4.6. Sprouts – a special problem

Seed sprouts, such as alfalfa and mung bean sprouts, represent a special problem in relation to microbial ecology. Sprouts are produced by first soaking viable seeds and then placing them in a warm humid environment to foster rapid germination. These are the same conditions that promote the growth of bacteria that may be present on the surface of the seed. This can result in a finished product that has high bacterial levels including, in some instances, pathogens (Splittstoesser et al., 1983; Andrews et al., 1982; Jaquette et al., 1996). For example, 57% of sprouting seeds taken from a retail market contained *B. cereus*, and upon germination, the levels reached 3.7–5.4$\log_{10}$ CFU/g (Harmon et al., 1987). En-

teric pathogens are occasionally isolated from sprouting seeds, however, *Staphylococcus aureus* isolations are more common (Andrews et al., 1979; O'Mahony et al., 1990; Prokopowich and Blank, 1991). The difficulty in controlling pathogen growth on sprouts is that treatments that inactivate bacteria also tend to decrease seed germination. Use of chlorinated water (1,000 ppm) in combination with strict sanitation are recommended as means for reducing bacterial levels (Brown and Oscroft, 1989), and 1,800–2,000 μg sodium hypochlorite/ml, 6% hydrogen peroxide, and 80% ethanol have been used experimentally to achieve a 1000-fold decrese in salmonellae levels (Beuchat, 1997). However, even after 10 min, these treatments were not able to eliminate the pathogen from seed surfaces. Recent investigations indicate that a 5-min exposure to moderately high temperatures (57–60°C), alone and in combination with 1000 ppm sodium hypochlorite, substantially reduced salmonellae on alfalfa and rice seeds (Jaquette et al., 1996; Piernas and Guiraud, 1997).

4.7. Hazard analysis critical control point (HACCP)

There have been few attempts to integrate the various steps associated with the production and processing of fresh produce into the development of farm-to-table HACCP systems. Two trade associations and a collaborative team from Clemson University, CDC, and The University of Georgia developed model HACCP programs for three specific commodities (sprouted seeds, International Sprout Growers Association; shredded lettuce, International Fresh-cut Produce Association; and tomatoes; Rushing et al., 1996), but complete validation of these plans has not yet been accomplished.

The NACMCF has long recognized that HACCP is the most effective and flexible system for assuring the microbiological safety of a variety of foods (NACMCF, 1998). The Committee anticipates that HACCP plans would be equally useful for fresh produce. However, currently available data are insufficient to develop validated HACCP plans for most fresh produce. Further, the requirements for prerequisite programs (such as GAPs and GMPs) that are the foundation on which HACCP systems are based are still being defined.

4.8. Control measures for fresh produce

To prevent foodborne diseases associated with fresh produce, it is necessary to prevent initial contamination, and reduce, eliminate, and prevent amplification of pathogens. Regulation of fresh produce is difficult because fruits and vegetables are simultaneously raw agricultural commodities and ready-to-eat foods. Proper sanitation at all levels in the fresh produce chain, from farm-to-table, is crucial. This includes the avoidance of the use of untreated manure as fertilizers, proper sanitary systems, and hand-washing facilities for the workers in the fields; the use of clean equipment and transportation vehicles; good hygiene in the processing facilities and in the kitchen; and measures to prevent cross-contamination. Persons with infections that can be transmitted through contamination of foods should not handle produce items until these workers are no longer infectious. As amplification points involving water are commonly implicated in fresh produce-associated outbreaks, the quality of irrigation water at the production level and the quality of water/ice used after harvesting is of prime importance. To minimize the growth of pathogens, it is important to maintain proper temperatures during processing, transportation, and storage of fresh produce; proper temperatures are commodity specific. For minimally processed fresh produce, the use of pasteurization, addition of growth inhibitors, or lowering of pH should be considered as barriers to bacterial survival and growth. Because *E. coli* O157:H7 and other pathogenic microorganisms survive well at pH 4.0, acidification cannot be generally relied on as the sole means of assuring the safety of fresh produce.

Research on methods for physical and chemical sanitation of fresh produce and studies on survival of pathogens from seed to fruits/vegetables, as well as determining survival and growth characteristics for likely pathogens in minimally processed fresh produce under retail conditions, need to be pursued. Prevention of foodborne illness is also an education issue. Education and information within the industry, at the retail level, and to the public should be undertaken. All growers, harvesters, processors, preparers, and consumers along the food chain from farm-to-table need to know, how to handle the products correctly, how to prevent cross-contamination, at what temperatures different foods are to be kept during storage, and how long processed produce can be kept before consumption. The fresh produce industry has recently begun implementing a Good Manufacturing Practices (GMP) program. This will help assure the safety as well as the quality of the fresh produce that reaches the market. However, it is important to remember that the problem of foodborne diseases is an international problem, especially when it comes to fresh produce. We cannot address this issue solely at a national level; international collaboration on food safety issues is crucial.

There is also a need for improved surveillance for foodborne illness. Regional and national surveillance systems are necessary for detection of diffuse clusters and for investigations of 'sporadic' cases. A rapid investigative response to clusters of cases, which is often crucial for successful investigations and product tracing, relies upon a well-functioning surveillance system. Subtyping (serotyping and/or molecular typing) of isolates is usually important for detection and investigations, and development and use of improved subtyping

techniques are critically needed in the public health laboratory network.

4.9. Microbial contamination of fresh produce

There are several steps in the fresh produce chain and many points for potential microbial contamination in each of these steps (Beuchat, 1996). Factors that may result in preharvest contamination of fresh produce include the use of manure as a fertilizer, fecal contamination from feral animals and employees, the use of contaminated irrigation water, the presence of feral or domestic animals, and human handling. At the postharvest level, critical factors include the use of contaminated wash water or ice, human handling, the presence of animals, the use of contaminated equipment or transportation vehicles, nonoptimal processing, cross-contamination, and improper storage, packaging, and display temperatures. Table 5 shows examples of bacterial pathogens that have been isolated from fresh produce.

Normally the exterior of the produce acts as a physical barrier, preventing bacteria from penetrating into the interior. However, the surface of produce is complex and difficult to clean, and pathogens may adhere tightly to this surface. Once surface integrity is broken, bacterial growth can be rapid. Processing of fresh produce increases the risk of bacterial contamination and growth. Cutting/shredding, juice processing, and heat treatment make produce more vulnerable to microbial adulteration.

4.10. Produce industry trends

Historically, fruits and vegetables have been used by those who grew them; what was possible to grow was severely limited by geography, climate, rainfall, season, and the ability to transport the produce to other places. With advances in plant breeding and genetics, improvement in agricultural practices, shelf-life extension technologies, temperature controls and distribution systems, there are now over 300 commodities offered for sale in produce departments of supermarkets throughout the US. Technological advances have expanded the global market, and produce is now available year round, nearly eliminating seasonality and providing consumers with a wide variety of products. The average number of commodities offered for sale in supermarket produce departments has increased from 65 items in 1975 to 340 items in 1995, a 432% increase.

With the increase in the number of produce items available for purchase in supermarkets, comes change in the packaging and display of fruits and vegetables within the produce department. Many items are sold in vacuum- or modified-atmosphere packaged containers. Supermarkets are now using water misters to keep produce items fresh. Many of these innovations yield environments in which microorganisms may flourish.

US consumers, motivated by increased interest in healthy diets, nutrition, and physical fitness have driven fruit and vegetable consumption to record levels. The total US per capita consumption increased 20% between 1970 and 1994. US per capita consumption of fruits and vegetables in 1970 was 225.6 and 222.3 pounds, respectively. These dramatic increases in fresh produce consumption have been due to better quality, increased variety, and year-round availability, while price, convenience, and variety have boosted vegetable consumption. US Department of Labor statistics indicate that consumers have increased their spending on produce relative to other foods.

Production of fruits and vegetables can vary considerably from year-to-year because of weather, planting and harvesting conditions, pests, etc., but production has been increasing. In 1994, fruit and vegetable production totaled 136.8 billion pounds, an increase of almost 61% from 85.0 billion pounds in 1970. Appendix B Table B1 provides an estimate of fresh fruit and vegetable sources and use of supply.

Table 5
Examples of bacterial pathogens isolated from fresh produce.

Country	Pathogen	Produce item	Reference
Egypt	*Salmonella, Shigella*	Salad greens	Saddik et al., 1985
United Kingdom	*Salmonella* Saint-paul	Bean sprouts	O'Mahony et al. 1990
Italy	*Salmonella*	Fennel, lettuce	Ercolani, 1976
	Yersinia enterocolitica	Vegetables	Gola et al., 1990
Netherlands	*Salmonella*	Cauliflower, endive, lettuce, eggplant, alfalfa sprouts, pepper	Tamminga et al., 1978
Spain	*Salmonella*	Vegetables	Garcia-Villanova Ruiz et al., 1987
Surinam	*Salmonella*	Chili	Tamminga et al. 1978
Thailand	*Salmonella*	Bean sprouts	Jerngklinchan and Sartanu, 1993
United States	*Campylobacter jejuni*	Mushrooms	Doyle and Schoeni, 1986
	Salmonella	Vegetables	Rude et al., 1984

Representative industry practices are further explained in Appendix B.

5. Research needs

5.1. Introduction

Many critical questions remain unanswered regarding the introduction and transmission of foodborne pathogens in fresh produce. Answers to these questions which can be derived through research are essential to develop effective methods for enhancing produce safety. Those measures which control pathogen introduction, persistence, growth, and transmission warrant the highest priority for research.

5.2. Control of pathogen introduction and persistence

The risk of illness associated with produce changes with time and place for the same pathogen-product combination. It also changes for different combinations in the same time and place. Therefore, it is unlikely that a single measure will provide equal control of all pathogens associated with a product or be applicable across a broad range of commodities. Interventions to prevent contamination, or decontaminate, or to prevent amplification can be designed for each component of the farm-to-fork chain, but their relative contributions to the overall control of pathogen transmission will vary. Priority for the allocation of resources and support should, therefore, be to those areas which will most efficiently and effectively control those produce-associated pathogens with the greatest impact on public health.

1. Growing and harvesting conditions and practices in agricultural operations in the United States and in countries exporting fresh fruits and vegetables to the US vary widely. An evaluation of various practices is needed to identify their impact on the safety of fresh produce.
   - The number and range of potential human pathogens that can be introduced during growing and harvesting may be a function of the source of contamination, including people, practices, and the environmental conditions. Examples of relevant research questions during growing and harvesting include the following:
     - What is the impact of worker hygienic practices during growing and harvesting (e.g., availability and use of field toilets and hand washing facilities) on the safety of fresh produce, and how can safety be improved?
     - What is the impact of growing and harvesting methods (eg., irrigation, fertilization, pest control) on produce safety?
     - Which environmental factors (e.g., other agricultural activities in the area, and indigenous flora and fauna, rainfall, temperature) have important negative or positive impact on produce safety?
   - Conditions and practices during growing and harvesting can influence the relative importance of various mechanisms through which external surfaces of fruits and vegetables become contaminated with human pathogens. Relevant research questions include the following:
     - Can conditions and practices be modified in ways that increase or decrease the potential for surface contamination?
     - Which mechanisms of internalization are important for various product–pathogen combinations under different conditions and practices of growing or harvesting?
     - Do bruises or niches in or on fruit and vegetable surfaces present an increased risk of contamination?
     - How should manure be treated, and applied to soil for fruit and vegetable production?
     - Do climate, soil conditions, and growing practices (e.g., type of irrigation) influence contamination?
2. Conditions and practices during handling of fresh produce also are important factors the introduction, amplification, and elimination of human pathogens. Activities that disrupt natural external barriers, alter existing microflora, and modify plant physiology among the factors that can increase or decrease risk of contamination with human pathogens. Relevant research for handling and processing includes the following:
   - Investigate the influence of various methods of chilling field-packed produce on the survival of human pathogens associated with produce.
   - Develop and evaluate the efficacy of cleaning/sanitizing methods to eliminate or reduce produce-borne human pathogens.
   - Evaluate and identify effective field hand-washing and disinfecting procedures used by produce pickers/handlers.
3. Human pathogens may be introduced or permitted to grow under certain conditions and practices of transport and distribution.
   - Research is needed to clarify the effects of various temperature/humidity combinations on survival and growth of important produce-associated pathogens.
   - The effect of various protective packaging conditions, including packaging atmospheres on the fate of pathogens needs to be determined.
4. Opportunities may exist to prevent the introduction of human pathogens, prevent their amplification, and eliminate or reduce those already present during marketing and preparation of fresh produce.

- Identify ways to modify conditions and practices during retail consumer sale to decrease the risk of produce-associated disease.
- Modify specific salad bar practices and equipment designs to improve the safety of fresh fruits and vegetables offered to consumers in salad bars.
- Identify ways that consumers and food workers can prevent the introduction of and can eliminate or reduce pathogens from fresh produce in food service and the home and retain product quality.

5.3. Control of pathogen transmission

Research is needed to elucidate the extent to which foodborne pathogens are transmitted by fresh produce and the methods for preventing foodborne pathogen transmission via fresh produce. Such research is listed in order of importance from the perspective of our immediate ability to prevent foodborne disease:

1. Given the multiple sources for possible contamination of fresh produce, and the fact that there are no recognized terminal critical control points for fresh fruit and vegetable production beyond pasteurization of fruit juices, there is an immediate need for identifying means for reducing pathogens on fresh produce while retaining sensory attributes desired by consumers.
2. Reliable assay systems which enable confirmation of the effectiveness of any type of pathogen reduction method used for fresh produce are needed. For some foodborne pathogens such as *Salmonella* and *E. coli* O157:H7, current methods for pathogen identification and viability testing are readily available. However, for other pathogens such as *Cyclospora*, research into the routine propagation and enumeration of these agents is needed.
3. The sensitivity and specificity of the current disease surveillance system in this country that is used to detect and explain the epidemiology of foodborne disease needs better definition. For example, the recent FOODNET system did not detect the cyclosporiasis outbreak associated with Guatemalan raspberries. In addition, research regarding the use of molecular subtyping of foodborne pathogens on a regional or national basis to detect outbreaks that are widely disseminated and of low-level contamination is critical to enable a rapid response.
4. The most effective way for conducting fresh produce tracebacks when outbreaks occur needs to be determined. Such tracebacks need to be comprehensive from the point of production to the consumer and they need to be rapid.
5. The most effective ways for conducting epidemiologic investigations and evaluating data from such investigations to enable accurate and timely identification of contaminated products need to be determined.
6. Better methods to sample, isolate and subtype foodborne pathogens from product and clinical specimens are needed. Isolation procedures with improved speed, sensitivity, and accuracy, and typing methods that are simple, inexpensive and discriminating would greatly benefit epidemiologic investigations. In addition, identifying nonpathogenic microorganisms that would serve as indicators of the presence of foodborne pathogens would be useful for product testing.

6. Recommendations

(1) Finding: Pathogenic bacteria and other infectious microorganisms can contaminate fresh produce during production, harvesting, packing, processing, distribution, marketing and preparation.

Recommendation: The Committee recommends that Good Agricultural Practices (GAPs) and Good Manufacturing Practices (GMPs) be developed to provide guidance on those agricultural and processing steps that can reduce the microbiological food safety risks associated with fresh produce. In the agricultural setting, GAPs should establish proper protocols related to water use, manure management, farmer/worker hygiene and transportation. GAPs and GMPs should be developed in a stepwise manner based on the risk associated with individual fruits and vegetables and the scientific data available.

(2) Finding: While the Committee recognizes that HACCP is the most effective and flexible means for assuring the microbiological safety of foods and food products, HACCP programs require detailed information on causes of contamination, the effects of production and processing practices on the prevalence and incidence of foodborne pathogens, and validated control measures that reduce or eliminate hazards (NACMCF, 1997). Currently, there are insufficient data to support the development of validated HACCP plans for fresh produce.

Recommendation: The appropriate government agencies, in consultation with industry, should support research initiatives that will provide data and technologies needed to develop effective HACCP programs that can be integrated with prerequisite programs based on recommended GAPs and GMPs.

(3) Finding: Despite current educational efforts, foodborne illness remains prevalent in the US. One reason is that people throughout the farm to table continuum may lack the knowledge about the hazards and risks involved and the skills related to safe food handling practices.

Recommendation: The Committee recommends that the appropriate federal agencies establish partnerships with the produce industry, academic institutions, states,

and consumer organizations to develop proactive and practical education and training programs to address the principles of safe food production, processing, distribution, preparation and storage of fresh produce. These educational efforts should be specifically designed for the following audiences:

- growers (field workers and packing house personnel)
- food processors (employees in juicing and fresh cut operations)
- transporters, distributors and wholesalers (individuals involved in the transportation and distribution of fresh produce)
- food retailers (associates working with produce in retail food stores)
- food service facilities (food service workers), and
- consumers

(4) Finding: There is a need to address food safety concerns related to fresh produce based on the risk they pose to public health. However, additional data are needed to conduct improved quantitative risk assessments.

Recommendation: The Committee recommends that federal agencies working through the Risk Assessment Consortium (The Risk Assessment Consortium was established in 1997 with interagency partners, HHS, USDA, and EPA, as part of the Joint Institute for Food Safety and Applied Nutrition, a collaborative activity of FDA's Center for Food Safety and Applied Nutrition and Center for Veterinary Medicine and the University of Maryland) identify additional information needed to conduct effective risk assessments of the microbial hazards associated with fresh produce. Working with industry and other interested parties, programs should be initiated to acquire the requisite data.

(5) Finding: When fresh produce has been implicated in foodborne outbreaks, the origin of the product, causes of contamination, and critical events leading to the outbreak have frequently not been identified.

Recommendation: The Committee recommends that investigative and regulatory agencies work with industry and others to develop better product identification and tracing systems. Outbreak investigations should identify causes of contamination and contributing factors whenever possible.

(6) Finding: Many deficiencies exist in our understanding of produce as a vehicle for foodborne disease, as well as in intervention strategies to prevent, eliminate or reduce pathogens on produce.

Recommendation: The Committee recommends that the research outlined in Section 5 of this document be initiated as soon as practically possible. Regulatory agencies should consider engaging in a public process for identifying the priority with which the research should be conducted.

(7) Finding: New technologies such as irradiation and other possible pasteurization processes, antimicrobial treatments, manure management methods, etc, may offer ways to mitigate or eliminate the microbial hazards associated with fresh produce. Many of these processes will require approvals from one or more federal agencies prior to their use. Delays in these approval processes could discourage, delay, or prevent research on the application of processes that could otherwise be providing additional public health protection.

Recommendation: The Committee recommends that FDA, USDA, and EPA eliminate from their evaluation processes those steps which unnecessarily impede timely use of technologies which address public health concerns associated with fresh produce.

Appendix A

See Tables 6–8

Table 6
Foodborne outbreaks due to fresh produce in 1973–87 and 1988–92 in the United States by specific etiology.[a,b]

Microorganism	1973–87[a]	1988–92[b]	Total
Salmonella sp.	9	9	18
Shigella sp.	3	0	3
Staphylococcus aureus	4	0	4
Bacillus cereus	3	0	3
Other bacteria	5	2	7
Giardia	1	2	3
Hepatitis A virus	1	4	5
Norwalk/like virus	3	0	3
Unknown	35 (55%)	23 (58%)	58 (56%)
Total	64	40	104

[a] Bean, N.H., & Griffin, P.M. (1990). Foodborne disease outbreaks in the United States, 1973–1987: Pathogens, vehicles, and trends. *J. Food Prot. 53*, 804–817

[b] CDC (1996). Surveillance for foodborne-disease outbreaks – United States, 1988–1992. *MMWR 45*. (no. SS-5).

Table 7
Food vehicles implicated in confirmed foodborne outbreaks in Minnesota, 1990–1996[a]

Vehicle	No. of outbreaks (%)
Fresh produce	37 (30)
Meat or poultry	19 (16)
Combination food[a]	18 (15)
Multiple foods	23 (19)
Other	21 (17)
Unknown	4 (3)
Total	122 (100)

[a] A single food item with many components (e.g., sandwich, casserole).

Table 8
Etiologic agents associated with fresh fruit and vegetable outbreaks in Minnesota, 1990–1996

Agent	No. of outbreaks (%)	No. of salad bars (%)
Norwalk-like virus	20 (54)	2 (25)
Salmonella spp,	6 (16)	4 (50)
Campylobacter	2 (5)	1 (13)
Hepatitis A virus	2 (5)	–
Clostridium perfringens	1 (3)	1 (13)
Escherichia coli O157:H7	1 (3)	–
Giardia	1 (3)	–
Shigella sonnei	1 (3)	–
Chemical	1 (3)	–
Unknown	2 (5)	–
Total	37 (100)	8 (100)

Appendix B

B.1. Industry's positions and views about approaches to enhancing food safety

The following is a summary of the activities of four organizations as outlined in presentations to the Fresh Produce Subcommittee on 25 July 1997.

B.2. United Fresh Fruit and Vegetable Association (United)

United noted that it is essential to present the risks associated with consumption of fresh produce within the context of benefits. Given the widely recognized fact that fresh fruits and vegetables are an important component of a healthy diet, we all must aim to assure that any efforts aimed at enhancing the safety of fresh fruits and vegetables do not jeopardize efforts to advocate their increased consumption.

United explained the numerous challenges the fresh produce industry faces in addressing food safety issues including environment dynamics, industry diversity, commodity perishability, emerging pathogens, and traceback.

In order to address risks associated with production and handling of fresh produce United recommends that guidelines be aimed at specific practices including water, manure, field, facility and worker sanitation, and transportation. Through the development of guidance that focuses on these areas, food safety issues can be properly addressed.

The fresh produce industry has developed a broad guidance document (*Industrywide Guidance to Minimize Microbiological Food Safety Risks for Produce*) to identify principal areas of concern and to provide a template for growers that scientific justification s to why each area identified in the guidance is important. United supports prevention-based food safety systems in the form of Good Agricultural Practices and Good Handling Practices. They stressed that using the term HACCP to describe food safety programs implemented to address minimizing pathogen contamination during production and handling of fresh produce is inappropriate because of the lack of science available. While they appreciate the prevention principles behind a HACCP system, Good Agricultural Practices and Good Handling Practices most accurately describe these programs at this time. Until the appropriate science is available to support the identification of critical control points and their corrective actions, HACCP is not appropriate to describe the food safety systems developed by the industry.

Cooperative extension specialists at many universities are working with the industry in the development of food safety programs, including Dr Susan Sumner at Virginia Polytechnic Institute, and Dr Trevor Suslow at the University of California-Davis, who both presented on behalf of United Dr Sumner shared her experience working with the tree fruit industry in Virginia in developing HACCP systems for juice production. Dr Suslow presented his evaluation of practices in the production of various fruits and vegetables in California.

The need for research to be conducted to identify the potential points of contamination during production and handing of produce and preventing contamination was emphasized. United also stated the need for risk assessment and research to be conducted in order to guide any recommendations to address food safety practices during production and handling of fresh produce. They also stressed that preventing foodborne outbreaks associated with fresh produce must focus on educational programs at all points in the food chain from farm to fork.

B.3. International Fresh-cut Produce Association/Western Growers Association

The International Fresh-cut Produce Association (IFPA) is an international trade association that provides technical expertise and represents 525 companies involved in the processing of fresh-cut produce or companies that provide equipment, supplies, or services to fresh-cut processors. Sixty-five percent of IFPA's members have sales below $5 million per year. Typical products include: broccoli, cabbage, carrots, cauliflower, celery, garlic, lettuce, melons, mushrooms, onions, peppers, pineapple, potatoes, spinach, squash, and tomatoes.

The IFPA was established in 1987. In 1986, the third edition of its '*Sanitation Guidelines*' was published and re-named '*Food Safety Guidelines for the Fresh-cut Product Industry*'. This document provides voluntary recommended guidelines on sanitary procedures for the fresh-cut industry. It contains suggested guidelines for designing a specific HACCP plan for a fresh-cut operation. The IFPA proposes seven CCPs. Three CCPs are: temperature control, chlorinated water, and a metal detector. Other food safety initiatives proposed include: establishing a plant and equipment sanitation plan as a prerequisite to HACCP, conducting employee GMP training and establishing GMP monitoring programs as another prerequisite to HACCP, supplier certification systems, coding and product recall with greater use of date coding for product recall and traceback, and third-party audits. In 1997, the IFPA published a Fresh-cut Produce Microbiology White Paper and the second edition of its '*Handling Guidelines*'. In a 1997 IFPA membership survey, 61% of respondents had implemented a verified HACCP plan.

The Western Growers Association (WGA) was established in 1926, and has 3100 grower/packer/shipper/processor members, primarily located in California and Arizona. Its members represent 54% of the total produce shipped in the United States.

The IFPA and WGA joined forces in December 1996 to convene a food safety initiative to develop food safety guidelines to minimize microbial problems associated with fresh produce. The Initiative cooperated and partnered with government representatives from the California Food and Drug Branch, California Department of Agriculture, Arizona Department of Agriculture, US Food and Drug Administration Regional Offices; US Department of Agriculture, and local county agriculture executives to develop the guidelines.

The Initiative targeted five main areas of potential microbiologic risk:

1. Water quality including irrigation water, postharvest process water, pesticide spray carriers, and hand washing water;
2. Worker hygiene at preharvest, harvest, postharvest cooling, packinghouse, and processing levels;
3. Manure management for those who use manure including effective sterilization, proper storage, and proper application intervals;
4. Packinghouse and processing plant sanitation including the facility environment and equipment; and
5. Establishment and maintenance of the cold chain from cooling, storage, processing, shipping, retail display, and consumer handling.

The IFPA/WGA First Edition of *Voluntary Food Safety Guidelines for Fresh Produce* was finalized in August 1997. It was presented first in draft and then in final form to the Fresh Produce Subcommittee at the 25 July 1997 and 12 August 1997 meetings, respectively. A copy of the Voluntary Food Safety Guidelines are attached to this reported as Appendix B.

B.4. Florida Fruit and Vegetable Association

The Florida Fruit and Vegetable Association (FFVA) is a grower-shipper organization. FFVA's basic premise is 'Consumers deserve the ability to purchase product that has been produced, distributed, and marketed in a manner which minimizes the risk of foodborne illness'. The factors that define the development of FFVA's efforts are:

- Crops produced in the natural environment cannot be expected to be free of microbiological agents.
- Every crop production and handling system has its own unique characteristics and must be evaluated separately; a broad-brush approach cannot be used. Irradiation appears to be a promising technology but is not amenable to all crops. Tomatoes, for example, turn black.
- The chain of commerce associated with fresh fruits and vegetables is extremely complex, has many links, and the potential for introduction or enhancement of microbial populations may be present at any point in the chain.

An FFVA – Environmental and Pest Management Division Subcommittee was established in June 1994. Development of a draft guidance document was completed in May 1997, and the first field test was held later that month. Carrots, leafy vegetables, and sweetcorn were examined using the checklist. Following the test, a revised draft guidance document was prepared, and generic mitigation guidelines will be prepared in late summer or early autumn. A final draft for approval by the FFVA membership is expected in late 1997.

The draft guidance document contains the following elements:

- Overview of the issues of concern
- Checklist worksheets (which may later be tailored to individual commodities)
- Generic risk mitigation information
- Appendix of supporting information

  Food Safety from Farm to Table: A National Food Safety Initiative to the President, May 1997.

  Good Manufacturing Practices in Manufacturing, Packaging, or Holding Human Food, 21 CFR Part 110.

  Good Manufacturing Practices: Fresh Broiler Products, National Broiler Council, Revised May 1992.

The guidance document is designed to stand alone and is expected to give FFVA members an essentially complete look at their processes.

Two main areas will be addressed in a decision flowchart:

1. Sources of microbiological contamination for agents associated with foodborne illness (e.g., water, ice, human contact).

2. Identification of points in the harvesting process during each phase of crop production where microbiological agents of concern may be introduced or populations of microbiological agents enhanced.

The assessment process will consist of two major elements:

1. An inventory of the steps in production, harvesting, packaging, and pre shipment practices that meet one or more of the following criteria:
- Represents a potential contamination source
- Serves as a point source inoculum for harvested produce
- Represents an existing sanitation or risk mitigation process
2. The crop and market profile for individual commodities:
- Description of the product and its end use
- Characterization of crop production practices, including proximity to animals and water
- Characterization of harvesting practices, e.g., mechanically picked, hand-picked, washed, etc.
- Characterization of packing practices
- Characterization of preshipment practices, e.g., storage practices, first- in/first-out, stored for shipment, cooling, etc.

Several points about traceback should be noted. In Florida, as in other states, produce destined for interstate commerce must be clearly marked as to the grower/shipper of the product. However, when the shipping container is placed into the distribution system all grower control over the identification can be lost. This is further compounded by the common practices of repacking certain produce items (i.e., tomatoes) in the typical distribution scheme. These repacked products may or may not go back into the original shipping container. Another common practice involves the reselling of used shipping cartons in the produce industry. This typically occurs at the terminal market or retail end of the chain, with the typical purchaser being local vendors that need inexpensive containers for local distribution (i.e., farmers markets, roadside stands, U-pick operations, etc).

There also is the matter of secondary distribution where product is purchased by a buying broker who then diverts the product in transit to different locations or markets than those indicated in the original shipping documents. As a result of different standards (or economics), a load of produce may be redirected once it has been invoiced at the shipping point. This increases the difficulty of using shipping documents during traceback efforts.

B.5. Taco Bell

Taco Bell has more than 7000 restaurants and is the fourth largest Quick Service Restaurant (QSR) in the US. It has used fresh produce for more than 10 years, employing fresh-cut produce almost exclusively. Taco Bell buys approximately 10% of all the iceberg lettuce used in the food industry and operates a closed system from supplier to distributor to its restaurants.

As far as Taco Bell is concerned, HACCP is mandatory for its suppliers. Taco Bell compels their suppliers to meet 10 or more requirements, among which are HACCP plans, pest control, water analysis, process control, document control, residue control, GMPs, metal detection policy, product standards, recall program, and trailer cleaning program. The company places a great deal of reliance on finished product specifications, including microbiological, chemical, and physical testing. Taco Bell deals only with the largest suppliers because the samller ones cannot generally meet their demands. Taco Bell also has specific process requirements and expects international suppliers to meet its standards. Details of selected requirements follow.

HACCP. This covers procurement, storage, washing, packaging, and post packing. At a minimum, the supplier must have CCPs to cover these areas.

Microbiological testing. Suppliers are asked to look for a wide range of pathogens to reinforce GMPs. Generally speaking, when the microbiological test results become available, the product has already been shipped, and in many cases, consumed. Produce is the one product that does not have the microbiological analyses completed before release. Testing is done at both the supplier level and in-house.

Pesticides. Records of application and monitoring are required.

Traceability. Taco Bell considers this to be critical, and suppliers are periodically asked to carry out mock recall programs. The latter have been found to be highly effective. In a mock recall, the supplier must identify within 2 h how much product was produced and where it was shipped. Each supplier must also provide a listing of the raw materials involved, the use-by date, the growers used, and appropriate lot numbers.

Process controls. The supplier must be able to monitor all items involved in the process, e.g., chlorine concentration, temperatures, washing, etc.

Water analysis. This is used as an indicator; testing is generally done on a weekly basis.

Distribution. Temperature is a critical factor throughout the process, and great emphasis is placed on the distribution system. Transfer from the distribution center to the restaurant is monitored, and product placement in the trailer is important. A loss of 1° means the loss of one day of shelf life.

Taco Bell attempts to tie it all together from the supplier standpoint. Taco Bell grades suppliers relative to which is best from among the many industries. Each year, there is at least one supplier that qualifies as 'Best of the Best' from the Fresh Cut Produce Industry, and

receives recognition by Taco Bell. Kentucky Fried Chicken and Pizza Hut are sister divisions of Taco Bell and use the same basic approach.

Taco Bell restaurants. A number of procedures are in place. Taco Bell has established product and workplace HACCP plans for all items. This is new for the QSR industry. A third-party auditor is used to monitor and assess all the restaurants with a HACCP-based program. Classifications are assigned, e.g., 'A' restaurant, 'D' restaurants, and each restaurant is told where improvement is needed. Also in place is a three-level manager certification program in food safety and sanitation. Managers are assessed in terms of how well their employees are trained.

Future activities. Attempts will be made to anticipate needs, deal with labor issues, and address product shelf-life considerations. Taco Bell believes that HACCP will ultimately become more automated via auditing with electronic systems and hopes to pursue that route in the future (Table 9), (Fig. 1).

Table 9
Estimates of fresh fruit and vegetable sources and use of supply in the US, early 1990s (millions of pounds)

Produce	Production	Imports	Exports	Total	Per capita (lbs)
Fresh fruit					
Citrus	8400	200	2200	6400	25.6
Bananas	10	6200	–	6210	24.8
Apples	6000	250	800	5450	21.8
Other fruits	6800	1350	500	7650	29.4
Total	21,210	8000	3500	25,710	101.6
Fresh vegetables					
Major vegetables	24,000	2800	1750	15,050	100.2
Other vegetables	5000	500	250	5250	21.0
Melons, all kinds	5000	1000	5700	5700	22.8
Irish potatoes	12,300	600	12,500	12,500	50.0
Sweet potatoes	1100	–	1100	1100	4.4
Total	68,610	12,900	6200	75,310	300.0

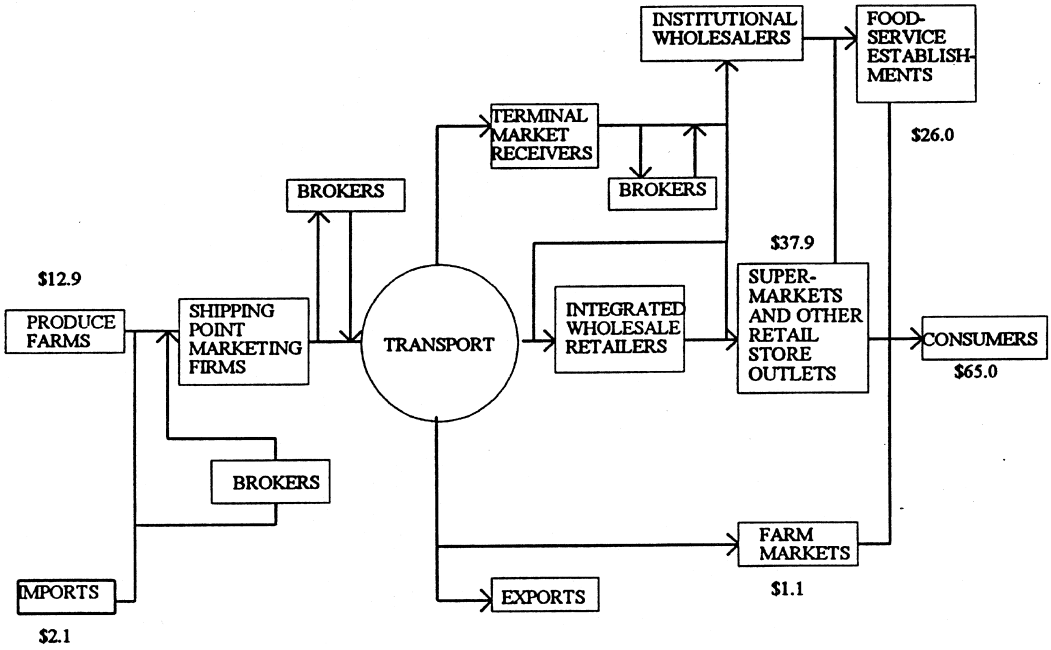

Fig. 1. The US fresh fruit and vegetable marketing system is a dynamic one that involves a complex network of growers, packers, brokers, importers, wholesalers, retailers, and the food service industry. Figure 1 describes this complex marketing system and provides the dollar amounts spent in selected areas of the system

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