

Critical factors affecting the microbial safety of fruits, vegetables and animal fresh produces: A review

Roshanlal Yadav¹, Bhawna Chugh²

^{1,2}Department of Food Technology, Bhaskaracharya College of Applied Sciences (University of Delhi), Delhi, India

ABSTRACT

Fruits and vegetables play a very significant role in nutrition and healthy diet, thus various initiatives have been undertaken by various countries to encourage consumers to eat more and more of fresh produce. This practice has led to an increased consumption, production and efficient distribution of raw fruits and vegetables. However, over past several years, unacceptable outbreaks of foodborne illness have been reported with this commodity. Contamination of agricultural produce may occur at any stage of a supply chain, from farm to fork that is harvesting, production, processing, storage and distribution but once they get contaminated it very difficult to sanitize them. Factors contributing to contamination includes changes in processing and agronomic practices, an increase in per capita consumption of raw fruits and vegetables, increased international trade and distribution, and an increase in the number of immuno-compromised consumers. The microbial load can be reduced by using conventional surface methods, but cannot eliminate pathogens completely. Electrolyzed water, Chlorine dioxide, cold atmospheric plasma, UV light, hydrogen peroxide, organic acids and acidified sodium chlorite show assurance, but irradiation at 1 kGy in high oxygen atmospheres proves to be the most effective method to assure elimination of both internal and surface contamination of produce by pathogens. Salmonella (tomatoes, seed sprouts and spices) and Escherichia coli O157:H7 on leafy greens (spinach and lettuce) are the pathogens of major concern nowadays. Factors responsible for illness are reviewed in this paper.

Keywords: Fresh produce, Pathogens, Foodborne illness, Salmonella Escherichia coli O157:H7.

INTRODUCTION

The importance of fresh produce in the diet is undisputed because of its nutrients, vitamins and fiber contribution. People's interest nowadays shifting towards the consumption of fresh produce for many reasons such as increased awareness towards healthy eating habits together with round year availability of them from the global market. Along with this, it has been reported that rates of foodborne illness are also increasing at alarming rate. This increase may be due to: changes in personal consumption, increased intensity of livestock production near areas of intense (extensive) produce production, greater availability of produce worldwide (some originating from countries with uncertain sanitary practices), and increased numbers of immuno-compromised consumers[1]. Studies have shown that Escherichia coli, Salmonella typhimurium, Listeria monocytogenes, Staphylococcus aureus and Enterococcus faecalis and other emerging food-borne pathogens are the common foodborne pathogens that can create a high health risk often with fatal consequences.

As people like to eat minimally processed or raw fresh produce, therefore, the presence of pathogens possess a serious threat and food safety becomes major concern for consumers. During production in the agricultural field, they can easily be contaminated from sources such as soil, water, air, birds, animals and insects. Further, cutting, slicing or peeling cause tissue damage which releases nutrients and facilitates the growth of microorganisms [2]. Following above sources, processing, packaging, transportation, retailing, storage, distribution and even handling at home can create conditions that can support microbial growth. Because of high water activity and nutrient load, fresh produce bear high growth of deadly pathogens. Serious outbreaks have been reported in Canada, USA, the UK, Japan and some other parts of the world

necessitating more effort for microbial food control[3]. Cleaning and sanitizing using effective sanitizer have been reported out as important components of the hygiene practices in a food and agricultural industry. During processing of fruits, vegetables, poultry, meat etc. chlorine rinses finds major application for pathogen control. Other various processes have been proposed as alternatives for eliminating or substantially decreasing bacterial populations.

These includes sodium bisulfite [4], sulfur dioxide[5], organic acids[6], calcium chloride[7 & 8], acidified sodium chlorite[9], and ozone[10]. However, chemical residues, discoloration of food produce where they are applied, limited effectiveness and elevated cost makes their narrow use in industries. Chlorine sanitizers, on the other hand, are therefore the most commonly used chemical sanitizer to date. Although chlorine sanitizers are popular and has been intensively used in food industry, increasing public health concerns about the possible formation of chlorinated organic compounds such as chloramines and trihalomethanes and the emergence of new more tolerant pathogens, have raised doubts in relation to the use of chlorine especially by the fresh-cut industry[11]. The prolonged exposure to chlorine vapors has been reported to result in the irritation to the skin and respiratory tract of workers [2]. Because of the lack of its safety and efficacy, use of chlorine will eventually bear restrictions by regulatory agencies, therefore, requires developing future alternatives. In 1998, FDA released a "Guide to Minimize Microbial Hazards for fresh fruits and vegetables", providing GAPs that growers, packers, shippers and retailers should implement to control contamination during their operations [12].

MICROBIAL SAFETY STATUS

Some of the common pathogens associated with produce are *E. coli* O157, *Salmonella* and *Listeria monocytogenes*. *E. coli* outbreaks have been reported from conventional and organic lettuce and salad greens (Strain- number of cases or outbreaks; O145 -26 cases and 12 hospitalizations, O157- 60 cases and 30 hospitalizations, O157-33 cases and 13 hospitalizations), sprouts (O26- 29 cases and seven hospitalizations, O121- 19 cases and seven hospitalizations), berries (O157- 15 cases and two deaths, O26 -five cases and one hospitalization), and melons (O157-nine and six cases in two outbreaks) [13]. Related to produce, in 2006, a multistate outbreak of *Salmonella* Typhimurium in tomatoes led to 183 cases of illness including 22 hospitalizations. A large outbreak in 2008 linked to pepper and tomatoes resulted in 1,500 illnesses, 308 hospitalizations, and two deaths [14]. Additional produce outbreaks have been linked to avocado (2007; 46 illnesses), blueberries (2009; 14 illnesses, 2010; six illnesses and one hospitalization), salads (multiple outbreaks and hospitalizations), cantaloupe (18 outbreaks resulting in multiple deaths), sprouts (1999; 112 illnesses, two hospitalizations), cucumber (2012; 49 illnesses, 14 hospitalizations), tomatoes (2004; 429 illnesses and 129 hospitalizations, 1998; 86 illnesses and three deaths), and watermelon (2008; 594 illnesses, 31 hospitalizations, and 2010; 17 illnesses, 11 hospitalizations) [15]. Produce has also been associated with listeriosis on several occasions in U. S. history.

Consumption of *Listeria* -contaminated celery resulted in 10 illnesses and five deaths in Texas, 2010 [15]. Sprouts have also been a vehicle of *Listeria* transmission in several cases, one in 2008 in which 20 individuals became ill, and another 3 in 2012 that resulted in six illnesses and one death [15]. *Listeria* was identified as the only microorganism (compared to *Salmonella* and *E. coli*) to be capable of growth on the surface of cantaloupe [16], which was the vehicle of one large outbreak in 2011. This outbreak spread across 28 U. S. states, resulting in 147 illnesses and 33 deaths, several of which were associated with pregnancy and/or newborns [17]. Produce-related outbreaks have also been linked to other bacterial pathogens; *Shigella* (carrots, lettuce, parsley, tomatoes, lemons, strawberries, and melons), *Bacillus* spp. (onion, sprouts, and potatoes), *Campylobacter* (pepper, lettuce, peas, watermelon, tomato, and spinach), *Clostridium* spp. (cabbage, mushrooms, onions, lettuce, and peppers), and *Staphylococcus aureus* (peppers, potato, salad greens).

Additionally, outbreaks have occurred as a result of parasites; *Cryptosporidium* (apple cider and salad greens), *Cyclospora* (berries, green beans, arugula, and peas), and *Giardia* (unspecified vegetables) as well as viruses; Hepatitis A (strawberries, green onions, tomato, and salad greens), Norovirus (melons, strawberries, salad greens, tomato, green beans, grapes, broccoli, cucumber, asparagus, and onion), and Norwalk (melons, salad greens, and celery) [18]. Others have also reported that extended exposure to outdoor conditions may cause organically raised farm animals to be more likely infected by *Salmonella* and *Campylobacter*[19].

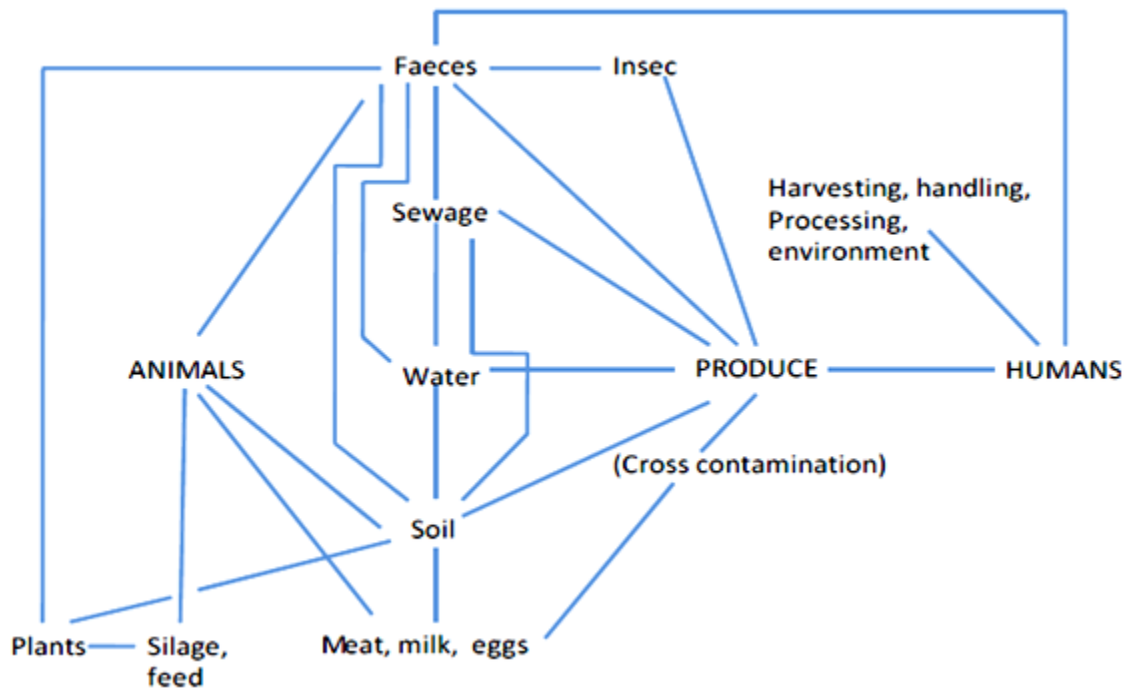


Figure 1. Sources by which produce can become contaminated with pathogenic microorganisms (adapted from [22]).

Organic meat production has the potential to have higher microbiological safety risks because of the strict restrictions on the use of pharmaceutical agents for therapeutic use (such as antimicrobials or parasiticides), raising the animals outdoors, use of slow - growing breeds and the smaller slaughtering facilities [20]. My research focused on evaluating the presence of some of the above-mentioned pathogens. The work in this article can be divided into two research goals. 1. Develop and study the food safety status of a model aquaponic system, and 2. Study the food safety status of a pasture - livestock agricultural system. Both the systems are modeled to be sustainable and do not employ pesticides, antibiotics or any other genetically modified organisms.

SOURCES OF PATHOGENS CONTAMINATING FRESH PRODUCE

In many fruits and vegetables, microorganisms find ideal conditions for their growth because of high nutrient content and neutral pH, especially in vegetables. Their structure consists of polysaccharides cellulose, pectin and hemicelluloses. With the help of extra cellular lytic cells, these spoilage microorganisms degrade above-mentioned polymers to release water and other intracellular materials to use them as nutrients for their growth. Although spoilage bacteria, mold and yeast predominate raw fruits and vegetables, isolation of pathogenic bacteria, parasites and viruses are not infrequent [21]. Factors that compromises the safety of fresh produce may occur either pre or post-harvest.

Beuchat [21] suggested a probable mechanism by which deadly microorganisms can contaminate fresh produce as shown in fig.1. Pre- harvest sources of contamination mainly includes soil, feces, irrigation water, water used to supply fungicides and insecticides, dust, insects, inadequately composed manure, wild and domestic animals, and human handling [22]. Human handling also contributes in post-harvest factors along with harvesting equipment, transport containers, wild and domestic animals, insects, dust, rinse water, ice, transport vehicles and processing equipment [23]. Pre-harvest contamination can occur directly or indirectly in the production chain. Direct sources comprises of untreated animal manure using as fertilizer, and wild lives bearing deadly pathogens. Practices such as irrigation with contaminated water, or contamination through run off water from nearby areas having faecal deposits, forms indirect sources of pre-harvest contamination.

The soil is natural and major reservoir for the variety of deadly micro flora such as *B. cereus*, *Cl. botulinum* and *Cl. perfringes*, *L. monocytogenes* and *Aeromonas* but this outline of pathogens get increased as livestock manure is added to the soil. Use of animal manure in agricultural practices gives easiest and most cost effective way to complete the nutrient requirements of crops and also to sustain fertility of the soil. Use of livestock manure is especially important in organic

plant production where mineral nitrogen fertilizers are banned. It has been reported that cattle is used as primary source of animal manure followed by a pig, chicken and horse manure to variable degrees. Livestock manure is a potential source of zoonotic pathogens such as Salmonella, Campylobacter and E.coli O157:H7 but their presence depends on the type of animal species and biological status of the herd. Under microbiologically favorable conditions, these zoonotic pathogens can stay for several hours even without input of any new manure. Faeces may naturally contain between 102 and 105 CFU/g E. coli and between 102 and 107 CFU/g Salmonella spp. [24], slurry between 10 and 104 CFU/g E. coli and Yersinia spp. [25] and manure between 102 and 107 CFU/g Salmonella spp. [26].

Also, *C. jejuni* is a normal member of the gastrointestinal micro flora of poultry, pigs, and cattle [27]. Although the presence of the specific pathogen is majorly decided by the animal source but the form of manure in terms of solid or slurry and their handling also affects the presence of the pathogen. Aeration of animal livestock and stirring of slurries promotes decomposition as well as helps to reduce pathogens. Similarly, temperature rise during composting helps to reduce pathogens, although this depends on the temperature reached [28]. Therefore, use of biosolids and manures involving solid manure, manure slurries, manure tea must be closely supervised to control potential of pathogen contamination. The government has established a number of guidelines on good agricultural practice (GAP) for fruit and vegetable production. Although these are not specific to organic produce but seems to be widely accepted agricultural practices. The Codex Alimentarius is a global reference point for ensuring the safety and quality of food that provides, among others, the code of hygiene practice for fresh fruits and vegetables (CAC/RCP53-2003, revised 2010) [28]. This codex provides a number of precautions related to manure use as a fertilizer, which is deliberated to control the microbial contamination.

An indirect way of pre-harvest contamination is the use of agricultural water (irrigation water) contaminated with animal or human fecal matter. Agricultural water refers to water used in the growing environment (for example, field, vineyard, or orchard) for agronomic reasons. It includes water used for irrigation, transpiration control (cooling), frost protection, or as a carrier for fertilizers and pesticides [12]. Water can be a transporter of many microorganisms involving pathogenic species of *Escherichia coli*, *Salmonella*, *Shigella*, *Vibrio cholera*, *Cryptosporidium parvum*, *Cyclospora cayentanensis*, *Giardia lamblia*, *Toxoplasma gondii*, and the Norwalk and hepatitis A viruses. The quality of irrigation water and type of irrigation system influence the microbial safety of fresh produce [29, 30, and 31].

Flood and spray irrigation represent the greatest risk because contaminated water can be directly deposited onto the edible leaves of produce [12]. Solomon et al. [32] studied the effect of irrigation types on the presence of *E. coli* O157:H7 and found that 90% of lettuce plants which had been spray-irrigated with water containing 7 log CFU/ml of *E. coli* O157:H7 were contaminated, while only 19% were contaminated when surface irrigation was used with the same concentration of *E. coli* O157:H7. Researches have shown that use of contaminated agricultural water can increase the frequency of pathogens in fresh produce. In 1990 and 1993, two major outbreaks, involving at least 300 cases in four states attributed to *Salmonella* species, were linked to consumption of fresh tomatoes [33 & 34]. Tomatoes from both outbreaks were detected and found that a water-bath in single packing facility was the likely source of contamination. Produce that involves large surface area (such as leafy vegetables) or foster attachment or entrapment may possess greater risk from pathogens especially if contact happens close to harvest or during the post-harvest time.

Factors such as cost and inconsistent availability of potable water can be cause for reuse of non-potable (gray or waste) water of unknown quality for irrigation, which may aggravate contamination of fresh produce. The WHO/FAO [35] recommended that the fecal coliform level of wastewater used for irrigation of fresh produce should not exceed 1000 CFU or MPN/100 ml. Several studies have reported that reconstituted pesticides may also act as potential sources of *Salmonella*, *Shigella*, *E. coli* O157:H7 and *L. monocytogenes* [36 & 37]. Ng et al. [37] tested 10 commercial pesticides (herbicides, insecticides and fungicides) and after reconstitution in sterile water to their suggested concentration, it was observed that two of the pesticides supported the survival as well as the growth of inoculated *Salmonella*, *Pseudomonas* and *E. coli* spp. Processing of fresh produce contains various steps and each step may be responsible for pathogen contamination. It involves human as well as mechanical contact, immersion in water, and cutting or slicing, which not only have the potential to contaminate produce with pathogens, but also can enhance bacterial growth [38 & 39]. During processing, protective barrier provided by the epidermis to control the introduction of microbes on the fruit surface is removed. Also, tissue damage occurs due to cutting, shredding and slicing which makes the fresh produce more susceptible towards microbial attack. Personal hygiene of farm workers is considered a dominant factor that influences shifting of pathogenic bacteria to fresh produce. Outbreaks of foodborne illness associated with fresh and minimally processed produce have been observed in past few years as a result of fecal contamination.

Therefore, an operator must ensure that all workers should follow agriculturally and management practices to minimize the direct or indirect contact between the fecal material and fresh produce. In addition, infectious diseases that involve boils, sores, or infected wounds can be a potential source of disease-causing microorganisms. The importance of food workers

understanding and practicing hygiene principles cannot be overstressed. They can unintentionally contaminate fresh produce, water supplies, and other workers, and transmit foodborne illness if they do not understand and follow basic hygienic principles [12]. For example, in 1994, there was a community hepatitis an outbreak in New York among individuals who had consumed bakery foods [40]. The source of the infection was a baker who contaminated baked goods while applying sugar glaze. In 1995, a foodborne outbreak, culture-confirmed for *Salmonella typhimurium*, occurred in a Minnesota nursing home [41]. Data from the investigation showed that the *Salmonella* was transferred by the consumption of mechanically softened foods, possibly contaminated by an infected employee.

Therefore, all personnel including even those indirectly involved in fresh produce operations, such as pest control operators, equipment operators and potential buyers follow established hygienic practices. At last, Cold storage inhibits the growth rate of most human pathogens. Therefore, fresh produce should be maintained below 50 C to reduce the proliferation of spoilage and pathogenic organisms [42]. It is proven that microbes of concern can survive for longer periods in environments where plants are grown for food use. Produce contamination is more likely to happen when crops are grown in soil that contains pathogens when soil is enriched with untreated liquid or solid manures, when contaminated water is used for agricultural practices or preparing pesticides (herbicides, insecticides and fungicides) for use on crops, and when worker is having the unhygienic practices.

POTENTIAL OUTBREAKS ASSOCIATED WITH FRESH PRODUCE

According to WHO, a foodborne disease outbreak can be defined as "The occurrence of two or more cases of a similar foodborne disease resulting from the ingestion of a common food". According to the Center for Disease Control and Preventions (CDC), the most common food-borne infections are caused by the bacteria *Campylobacter*, *Salmonella*, and *E. coli* O157:H7, and by a group of viruses called calicivirus, also known as the Norwalk and Norwalk-like viruses. It is observed that *Salmonella* and *E. coli* O157:H7 continuously cause large outbreaks of foodborne illness linked to fresh produce [43, 12 & 31]. The incidence of foodborne outbreaks caused by contaminated fresh fruit and vegetables has increased in recent years [44]. Fresh produce like cilantro, cantaloupes, cucumbers, and peppers that are usually eaten raw cause more foodborne illness than any other single commodity of food, according to a study by the Center for Science in the Public Interest (CSIP). In the last 10 years, fresh produce caused 629 outbreaks and almost 20,000 illnesses. In 2012, 831 foodborne outbreaks were reported to CDC.

They were caused by a variety of pathogens, and 106 of them were confirmed *Salmonella*. *Salmonella* accounted for the most hospitalizations (64%) in outbreaks with a confirmed cause. In the largest recent outbreak, between March 2013 and July 2014, over 600 individuals in 29 states and Puerto Rico were infected with seven outbreak strains of *Salmonella* Heidelberg. This outbreak was associated with one brand of chicken that led to a company recall of over 40,000 pounds of chicken products and ended after the company instituted new control measures to reduce contamination. On December 2016, The U.S. Food and Drug Administration and the Centers for Disease Control and Prevention investigated an outbreak of hepatitis A illnesses linked to frozen Egyptian strawberries in smoothies served at Tropical Smoothie Café locations. As per CDC reports 134 people with hepatitis A linked to this outbreak have been reported from 9 states. Among people with available information, 52 people have been hospitalized.

The CDC reports that nine people infected with the outbreak strains of *Listeria monocytogenes* have been reported from four states (California, Connecticut, Maryland, and Washington) from September 2013 to May 2016. Epidemiology and laboratory evidence indicated that frozen vegetables produced by CRF Frozen Foods of Pasco, Washington, and sold under various brand names are one likely source of illnesses in this outbreak. The FDA facilitated the recall of at least 456 products related to this outbreak. Listeriosis can be fatal, especially in certain high-risk groups. These groups include the elderly, and people with weakened immune systems and certain chronic medical conditions (such as cancer). In pregnant women, listeriosis can cause miscarriage, stillbirth, premature labor, and serious illness or death in newborn babies. Fresh produce and sprouts have been implicated in a number of documented outbreaks of illness in countries such as Japan and EU. In the U.S. between 1995 and 1998, there were nine outbreaks of foodborne illness caused by *Salmonella* or *E. coli* O157:H7 due to consumption of fresh vegetable sprouts [45].

These outbreaks involved more than 1234 cases in Missouri, Michigan, California, Washington, Arizona, and Nevada, and in most cases, alfalfa or clover seed were implicated as the initial inoculum source. *E. coli* O157 outbreaks have been linked to apple cider, lettuce, radish, alfalfa sprouts and other mixed salads [1]. The largest *E. coli* O157:H7 outbreak occurred in 1996 in Japan which involved more than 12,000 cases with 12 deaths, and these were linked to the consumption of raw radish sprouts [46]. The FDA, the U.S. Department of Agriculture's Food Safety and Inspection Service (FSIS), the Centers for Disease Control and Prevention (CDC) investigated a multi-state outbreak of *E. coli* O157:H7 illnesses linked to ready-to-eat salads. On December 11, 2013, the CDC reported a total of 33 persons infected with the outbreak strain of *E. coli*

O157:H7 were reported from four states: Arizona, California, Texas, and Washington. Understanding the causes for the increasing contribution of contaminated produce to the overall number of foodborne illness will provide light on measures that are most effective to return this trend.

PRODUCED PATHOGEN

Escherichia coli

A large number of fatal multi - state outbreaks have been reported in fresh produce over many years. Produce is a prominent vehicle for E. coli transmission, accounting for approximately 20% of all produce outbreaks [47]. E. coli outbreaks have been reported from lettuce and salad greens (S train - number of cases or outbreaks; O145 - 26 cases and 12 hospitalizations, O157 - 60 cases and 30 hospitalizations, O157-33 cases and 13 hospitalizations), sprouts (O26 - 29 cases and seven hospitalizations, O121 - 19 cases and seven hospitalizations), berries (O157 - 15 cases and two deaths, O26 - five cases and one hospitalization), and melons (O157 - nine and six cases in two outbreaks) [13 & 15]. In a case of sprout, the source of the outbreak was traced back to the seeds. However, the source of contamination in case of other breakouts could not be identified. As many as 400 Shiga toxin-producing Escherichia coli (STEC) are known to exist but not all have been identified as causing human illness and not all cause human disease in the same severity [48]. The STEC strain most commonly associated with severe forms of the disease is E. coli O157:H7 , but it is not the only STEC known to cause disease.

In fact, at least 60 strains of STEC have 15 been linked to human illness worldwide [49], and a U. S. study completed by the CDC between 1983 and 2003 demonstrated as many as 14 different serogroups were implicated in human disease resulting from E. coli infection, in addition to illnesses that resulted in undetermined serotypes [50]. However, the same study demonstrated that approximately 70% of the infections caused by non - O157 STEC infections, that could be serotyped, were attributed to only 6 serotypes: O145, O121, O 111, O103, O45, and O26, which have been identified by the CDC and USDA - FSIS as the —Big 6l non - O157 STEC [51]. Non - O157 STEC is of major concern in many areas of the world. Some European countries report that over one-half of confirmed STEC infections are caused by non - O157 STEC [52]. It has been estimated that E. coli O157:H7 strains cause two - thirds of all E. coli human infection cases in the U. S., while non - O157 strains are responsible for the remaining cases [53]. When compared to E. coli O157:H7 and other enteric pathogens, non -O157 STEC are infrequently isolated and implicated in foodborne illness outbreaks. It is believed that this group of pathogens is largely under-accounted for, presumably due to ineffective laboratory screening and culturing methods [54].

SALMONELLA

Salmonella spp. have been implicated in outbreaks linked to produce; sprouts, tomatoes, cantaloupe, spinach, peppers, papaya, beets, cabbage, cauliflower, on an ion, and 16 let tuce [18]. In addition to animal products, nuts and produce have also been often implicated as vehicles for the transmission of Salmonella. Outbreaks with peanuts include one in 2006 when 715 became ill and 129 were hospitalized, in 2008 were 714 illnesses, 166 hospitalizations, and nine deaths, and also in 2012 when 42 became ill and ten were hospitalized [14]. A large outbreak in 2008 linked to pepper and tomatoes resulted in 1,500 illnesses, 308 hospitalizations, and two deaths [15]. Additional produce outbreaks have been linked to avocado (2007; 46 illnesses), blueberries (2009; 14 illnesses, 2010; six illnesses and one hospitalization), salads (multiple outbreaks and hospitalizations), cantaloupe (18 outbreaks resulting in multiple deaths), sprouts (1999; 112 illnesses, two hospitalizations), cucumber (2012; 49 illnesses, 14 hospitalizations), tomatoes (2004; 429 illnesses and 129 hospitalizations, 1998; 86 illnesses and three deaths), and watermelon (2008; 594 illnesses, 31 hospitalizations, and 2010; 17 illnesses, 11 hospitalizations) [14].

Though the outbreaks could be traced back to the firm/company producing the product, the actual source of outbreaks has not been identified. Salmonella spp. is a common inhabitant of the intestinal tract of birds, reptiles, mammals (livestock such as pigs and cattle, and humans), and insects [55]. It is therefore naturally secreted in feces and transmitted by water, plants, and soil. Once contracting Salmonella, a person or animal can become a carrier; shedding the organism in its feces without showing symptoms of the disease to infect others. Much research has indicated that Salmonella is capable of survival in the environment for extended periods of time. A prevalence study by Jensen et al. [56] demonstrated the ability of Salmonella to persist in a hog production environment for up to five (soil) or seven (shelters) weeks, and to be present in water associated with production. It has also been shown to survive in manure and manure -amended soils for up to 184 and 332 days, respectively [57]. Transfer of Salmonellaentericaserovar Typhimurium onto the surface of produce (carrot and radish) was shown when contaminated manure and irrigation water was applied throughout the growing season [58]. Yang et al. [59] demonstrated the ability of Salmonella Typhimurium to persist in chilled chicken processing water after

treatment with chlorine, and Mezrioui et al. [60] found Salmonella to have better survival rates in treated sewage water than that of E. coli.

LISTERIA MONOCYTOGENES

Produce has also been associated with listeriosis on several occasions in U. S. history. Consumption of Listeria-contaminated celery resulted in 10 illnesses and five deaths in Texas, 2010 [14]. Sprouts have also been a vehicle of Listeria transmission in several cases, one in 2008 in which 20 individuals became ill, and another in 2012 that resulted in six illnesses and one death [14]. Listeria was identified as the only microorganism (compared to Salmonella and E. coli) to be capable of growth on the surface of cantaloupe [16], which was the vehicle of one large outbreak in 2011. This outbreak spread across 28 U. S. states, resulting in 147 illnesses 18 and 33 deaths, several of which were associated with pregnancy and/or newborns [15]. Being present in natural environments, unsanitary conditions and poor equipment maintenance are major causes of Listeria contamination. Listeria monocytogenes is naturally found in the environment (such as soil), making it an easy transfer onto growing produce, especially root crops. Listeria monocytogenes has been linked to outbreaks associated with the consumption of celery, sprouts, cabbage, cucumber, lettuce, tomato, and cantaloupe [18 & 14]. Listeria monocytogenes is widely found in the environment; present in the soil, vegetative material, fecal and water samples. Prevalence studies have found L. monocytogenes in raw milk, soft cheeses, meat, poultry, seafood, and fruit and vegetables [55]. A large number of mammals host the bacteria, including cattle, sheep, goats, and humans, as well as poultry and other fowl, ticks, flies, fish, and crustaceans [61]. The high prevalence of Listeria in the environment, combined with its ability to grow at low temperatures and readily form biofilms [62] leads to high concern over this pathogen in food processing environments. Persistence of some 19 strains within these processing environments [63] has led to zero tolerance for this pathogen in ready to eat (RTE) products [55].

OTHER PATHOGENS

Produce-related outbreaks have also been linked to other bacterial pathogens; Shigella (carrots, lettuce, parsley, tomatoes, lemons, strawberries, and melons), Bacillus spp. (onion, sprouts, and potatoes), Campylobacter (pepper, lettuce, peas, watermelon, tomato, and spinach), Clostridium spp. (cabbage, mushrooms, onions, lettuce, and peppers), and Staphylococcus aureus (peppers, potato, salad greens). Additionally, outbreaks have occurred as a result of parasites; Cryptosporidium (apple cider and salad greens), Cyclospora (berries, green beans, arugula, and peas), and Giardia (unspecified vegetables) as well as viruses; Hepatitis A (strawberries, green onions, tomato, and salad greens), Norovirus (melons, strawberries, salad greens, tomato, green beans, grapes, broccoli, cucumber, asparagus, and onion), and Norwalk (melons, salad greens, and celery) [18].

REFERENCES

- [1]. Beuchat, L. R. (2002), "Ecological factors influencing survival and growth of human pathogens on raw fruits and vegetables," *Microbes and infection*, 4(4), 413-423.
- [2]. Harris, L. J., Farber, J. N., Beuchat, L. R., Parish, M. E., Suslow, T. V., Garrett, E. H., & Busta, F. F. (2003), "Outbreaks associated with fresh produce: incidence, growth, and survival of pathogens in fresh and fresh-cut produce," *Comprehensive reviews in food science and food safety*, 2(s1), 78-141.
- [3]. Issa-Zacharia, A., Kamitani, Y., Muhimbula, H. S., & Ndabikunze, B. K. (2010), "A review of microbiological safety of fruits and vegetables and the introduction of electrolyzed water as an alternative to sodium hypochlorite solution," *African Journal of Food Science*, 4(13), 778-789.
- [4]. Krahn, T. (1977), "Improving the keeping quality of cut head lettuce," In *Symposium on Vegetable Storage* 62 (pp. 79-92).
- [5]. Bolin HR, Stafford AE, King AD Jr, Huxsoll, CC (1977), "Factors affecting the storage stability of shredded lettuce," *Journal of Food Science*, 42: 1319- 1321.
- [6]. Adams, M. R., Hartley, A. D., & Cox, L. J. (1989), "Factors affecting the efficacy of washing procedures used in the production of prepared salads," *Food Microbiology*, 6(2), 69-77.
- [7]. Izumi H, Watada AE (1995), "Calcium treatment to maintain quality of zucchini squash slices," *Journal of food science*, 60: 789-793.
- [8]. Izumi H, Watada AE (1994), "Calcium treatments affect storage quality of shredded carrots," *Journal of food science*, 59: 106-109.
- [9]. Allende A, McEvoy J, Tao Y, Luo Y (2009), "Antimicrobial effect of acidified sodium chlorite, sodium chlorite, sodium hypochlorite, and citric acid on Escherichia coli O157:H7 and natural microflora of fresh-cut cilantro," *Food Control*, 20: 23-234.
- [10]. Nagashima T, Kamoi I (1997), "Sterilization and preservation of vegetables by ozonated water treatment," *Food Preservation Science*, 23: 127-131.
- [11]. Singh, N., Singh, R. K., Bhunia, A. K., & Strohshine, R. L. (2002), "Effect of inoculation and washing methods on the efficacy of different sanitizers against Escherichia coli O157: H7 on lettuce," *Food Microbiology*, 19(2), 183-193.

- [12]. FDA (1998), "Guide to minimize microbial food safety hazards for fresh fruits and vegetables. Guidance for Industry," Food and Drug Administration, Center for Food Safety and Applied Nutrition. 1998 Oct 26.
- [13]. Danyluk, M. D., R. McEgan, A. N. Turner, and K. R. Schneider. (2014), "Outbreaks of foodborne illness associated with melons," University of Florida, IFAS Extension, FSHN14-11. Accessed on: 1 April, 2014.
- [14]. Center for Disease Control and Prevention (2011), "Investigation of outbreak of infections caused by Salmonella Saintpaul," Accessed on: 20 December, 2014.
- [15]. Centers for Disease Control and Prevention (2012), "Foodborne outbreak online database (FOOD)," Accessed on: 20 June 2014. Available at: <http://www.cdc.gov/foodborneoutbreaks/Default.aspx>.
- [16]. Behrsing, J., J. Jaeger, F. Horlock, N. Kita, P. Franz, and R. Premier (2003), "Survival of *Listeria innocua*, *Salmonella salford* and *Escherichia coli* on the surface of fruit with inedible skins," *Postharvest Biology and Technology*, 29(3): 249-256.
- [17]. Center for Disease Control and Prevention. (2012b), "Multistate outbreak of Listeriosis linked to whole cantaloupes from Jensen Farms, Colorado," Accessed on: 8 October, 2014.
- [18]. Olaimat, A. N., and R.A. Holley (2012), "Factors influencing the microbial safety of fresh produce: a review," *Food Microbiology*, 32(1): 1-19.
- [19]. Lund, V. (2006), "Natural living: a precondition for animal welfare in organic farming," *Livestock Science*, 100 (2-3): 71-83.
- [20]. Doyle, M.P., F.F. Busta, B.R. Cords, P.M. Davidson, J. Hawke, H.S. Hurd, R.E. Isaacson, K. Matthews, J. Maurer, and J. Meng. (2006), "Antimicrobial resistance: implications for the food system," *Comprehensive Reviews in Food Science and Food Safety*, 5(3): 71-137.
- [21]. Beuchat, L.R. (1998), "Surface Decontamination of Fruits and Vegetables Eaten Raw: A Review, Food Safety Issues, Food Safety Unit. World Health Organization, Geneva," WHO/FSF/FOS/98.2. Available at: http://www.who.int/foodsafety/publications/fs_management/en/surface_decon.pdf (accessed 19.08.11).
- [22]. Beuchat, L.R. (1996), "Pathogenic microorganisms associated with fresh produce," *Journal of Food Protection*, 59: 204-216.
- [23]. Burnett SL, Beuchat LR (2001), "Human pathogens associated with raw produce and unpasteurized juices, and difficulties in contamination," *Journal of Industrial Microbiology and Biotechnology*, 27: 104-110.
- [24]. Himathongkham, S., Bahari, S., Riemann, H., & Cliver, D. (1999), "Survival of *Escherichia coli* O157: H7 and *Salmonella typhimurium* in cow manure and cow manure slurry," *FEMS Microbiology Letters*, 178(2), 251-257.
- [25]. Kearney, T. E., Larkin, M. J., & Levett, P. N. (1993), "The effect of slurry storage and anaerobic digestion on survival of pathogenic bacteria," *Journal of Applied Bacteriology*, 74(1), 86-93.
- [26]. Pell, A. N. (1997), "Manure and microbes: public and animal health problem," *Journal of Dairy Science*, 80(10), 2673-2681.
- [27]. Warriner, K., & Namvar, A. (2010), "The tricks learnt by human enteric pathogens from phytopathogens to persist within the plant environment," *Current Opinion in Biotechnology*, 21(2), 131-136.
- [28]. Hoorfar, J. (Ed.). (2014), "Global safety of fresh produce: A handbook of best practice, innovative commercial solutions and case studies," Woodhead Publishing.
- [29]. Aruscavage, D., Lee, K., Miller, S., LeJeune, J.T. (2006), "Interactions affecting the proliferation and control of human pathogens on edible plants," *Journal of Food Sciences*, 71, R89eR99.
- [30]. Brackett, R.E. (1999), "Incidence, contributing factors, and control of bacterial pathogens in produce. *Postharvest Biology and Technology*, 15, 305e311.
- [31]. Warriner, K., Huber, A., Namvar, A., Fan, W., Dunfield, K. (2009), "Recent advances in the microbial safety of fresh fruits and vegetables," *Advances in Food and Nutrition Research*, 57, 155e208.
- [32]. Solomon, E. B., Yaron, S., & Matthews, K. R. (2002), "Transmission of *Escherichia coli* O157: H7 from contaminated manure and irrigation water to lettuce plant tissue and its subsequent internalization," *Applied and Environmental Microbiology*, 68(1), 397-400.
- [33]. Wood, R.C., C. Hedburg, and K. White (1991), "A multistate outbreak of *Salmonella javiana* associated with raw tomatoes," *Epidemic Intelligence Service*, 40th Annual Conference, CDC, Atlanta, GA, 1991.
- [34]. CDC, "Multistate outbreak of *Salmonella* serotype Montevideo infections," *EPI-AID* 93-79, 1993.
- [35]. World Health Organization/Food and Agriculture Organization (WHO/FAO) (2006), "Guidelines for the Safe Use of Wastewater, Excreta and Grey Water," *Wastewater Use in Agriculture*, vol. 2. Geneva, Switzerland. Available at: http://whqlibdoc.who.int/publications/2006/9241546832_eng.pdf (accessed 11.11.11).
- [36]. Guan, T.Y., Blank, G., Holley, R.A. (2005), "Survival of pathogenic bacteria in pesticide solutions and treated tomato plants," *Journal of Food Protection*, 68, 296e304.
- [37]. Ng, P.J., Fleet, G.H., Heard, G.M. (2005), "Pesticides as a source of microbial contamination of salad vegetables," *International Journal of Food Microbiology*, 101, 237e250.
- [38]. Brackett, R.E. (1999), "Incidence, contributing factors, and control of bacterial pathogens in produce," *Postharvest Biology and Technology*, 15, 305e311.
- [39]. Doyle, M.P., Erickson, M.C. (2008), "Summer meeting 2007: the problems with fresh produce: an overview," *Journal of Applied Microbiology*, 105, 317e330.
- [40]. Weltman, A.C., N.M. Bennett, D.A. Ackman et al. (1996), "An outbreak of hepatitis A associated with a bakery, New York, 1994: The West Branch, Michigan outbreak repeated," *Epidemiology Infection*, 117: 333-341.
- [41]. Minnesota Department of Health, *Foodborne and Waterborne Outbreak Summary, 1995*. Minneapolis, Minnesota.
- [42]. Rediers, H., Claes, M., Peeters, L., & Willems, K. A. (2009), "Evaluation of the cold chain of fresh-cut endive from farmer to plate," *Postharvest Biology and Technology*, 51(2), 257-262.
- [43]. Buck, J.W., Walcott, R.R., Beuchat, L.R. (2003), "Recent trends in microbiological safety of fruits and vegetables," *Plant Health Progress*, doi: 10.1094/PHP-2003-0121-01-RV

- [44]. Mukherjee, A., Speh, D., Jones, A. T., Buesing, K. M., & Diez-Gonzalez, F. (2006), "Longitudinal microbiological survey of fresh produce grown by farmers in the upper Midwest," *Journal of Food Protection*, 69(8), 1928-1936.
- [45]. Buchanan, R. L. (1997), "National Advisory Committee on Microbiological Criteria for Foods," *Journal of Food Protection*, 60(11), 1417-1419.
- [46]. Michino, H., Araki, K., Minami, S., Takaya, S., Sakai, N., Miyazaki, M., & Yanagawa, H. (1999), "Massive outbreak of *Escherichia coli* O157:H7 infection in schoolchildren in Sakai City, Japan, associated with consumption of white radish sprouts," *American journal of epidemiology*, 150(8), 787-796.
- [47]. Rangel, J.M., P.H. Sparling, C. Crowe, P.M. Griffin, and D.L. Swerdlow (2005), "Epidemiology of *Escherichia coli* O157:H7 Outbreaks, United States, 1982–2002," *Emerging Infectious Diseases* 11(4):603-609.
- [48]. Liu, D. (2010), "Molecular detection of foodborne pathogens. Florida," Taylor & Francis Group, LLC, Print.
- [49]. Bettelheim, K. A. (2003), "Non-O157 Verotoxin-producing *Escherichia coli*: a problem, paradox, and paradigm," *Experimental Biology and Medicine*, 228(4): 333-344.
- [50]. Brooks, J. T., E. G. Sowers, J. G. Wells, K. D. Greene, P. M. Griffin, R. M. Hoekstra, and N. A. Strockbine (2005), "Non-O157 Shiga toxin-producing *Escherichia coli* infections in the United States, 1983-2002," *Journal of Infectious Disease*, 192(8): 1422-1429.
- [51]. *FEMS Microbiology Letter* 282: 124-131, "United States Department of Agriculture Food Safety Inspection Service (USDA-FSIS)," Office of Policy and Program Development, Office of Public Health Science. (2010).
- [52]. Monaghan, A., B. Byrne, S. Fanning, T. Sweeney, D. McDowell, and D. J. Bolton (2011), "Serotypes and virulence profiles of non-O157 Shiga toxin-producing *Escherichia coli* isolates from bovine farms," *Applied and Environmental Microbiology*, 77: 8662-8668.
- [53]. Mead, P. S., L. Slutsker, V. Dietz, L. F. McCaig, J. S. Bresee, C. Shapiro, P. M. Griffin, and R. V. Tauxe (1999), "Food-related illness and death in the United States," *Emerging Infectious Disease*, 5: 607-625.
- [54]. Possé, B., L. DeZutter, M. Heyndrickx, and L. Herman (2008), "Novel differential and confirmation plating media for Shiga toxin-producing *Escherichia coli* serotypes O26, O103, O111, O145, and sorbitol-positive and negative O157,"
- [55]. Jay, J. M., M. J. Loessner, D. A. Golden (2005), "Modern Food Microbiology," Print. Springer Media, Inc. New York, NY.
- [56]. Jensen, A. N., A. Dalsgaard, A. Stockmarr, E. M. Nielsen, and D. L. Baggesen (2006), "Survival and transmission of *Salmonella entericaserovar* Typhimurium in an outdoor organic pig farming environment," *Applied Environmental Microbiology*, 72(3): 1833-1842.
- [57]. You, Y., S. C. Rankin, H. W. Aceto, C. E. Benson, J. D. Toth, and Z. Dou. (2006), "Survival of *Salmonella entericaserovar* Newport in manure and manure-amended soils," *Applied Environmental Microbiology*, 72(9): 5777-5783.
- [58]. Islam, M., J. Morgan, M. P. Doyle, S. C. Phatak, P. Millner, and X. Jiang. (2004), "Fate of *Salmonella entericaserovar* Typhimurium on carrots and radishes grown in fields treated with contaminated manure composts or irrigation water," *Applied Environmental Microbiology*, 70(4): 2497-2502.
- [59]. Yang, H., Y. Li, and M. G. Johnson (2001), "Survival and death of *Salmonella* Typhimurium and *Campylobacter jejuni* in processing water and on chicken skin during poultry scalding and chilling," *Journal of Food Protection*, 64(6): 770-776.
- [60]. Mezrioui, N., B. Baleux, and M. Troussellier (1995), "A microcosm study of the survival of *Escherichia coli* and *Salmonella typhimurium* in brackish water," *Water Research*, 29(2): 459-465.
- [61]. Gray, M. L., and A. H. Killinger (1966), "*Listeria monocytogenes* and listeria infections," *Bacteriological Reviews*, 30(2): 309-382.
- [62]. Valderrama, W. B., and C. N. Cutter (2013), "An ecological perspective of *Listeria monocytogenes* biofilms in food processing facilities," *Critical Reviews in Food Science and Nutrition*, 53(8): 801-817.
- [63]. Holch, A., K. Webb, O. Lukjancenko, D. Ussery, B. M. Rosenthal, and L. Gram. (2013), "Genome sequencing identifies two nearly unchanged strains of persistent *Listeria monocytogenes* isolated at two different fish processing plants sampled 6 years apart," *Applied and Environmental Microbiology* 79 (9): 2944-2951.