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Review

Microbiology of organic and conventionally grown fresh produce

Daniele F. Maffei[a,*], Erika Y. Batalha[a], Mariza Landgraf[a], Donald W. Schaffner[b], Bernadette D.G.M. Franco[a]

[a] Universidade de São Paulo, Faculdade de Ciências Farmacêuticas, Departamento de Alimentos e Nutrição Experimental, São Paulo, SP, Brazil

[b] Rutgers University, School of Biological and Environmental Sciences, Department of Food Science, New Brunswick, NJ, USA

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ABSTRACT

Fresh produce is a generalized term for a group of farm-produced crops, including fruits and vegetables. Organic agriculture has been on the rise and attracting the attention of the food production sector, since it uses eco-agricultural principles that are ostensibly environmentally-friendly and provides products potentially free from the residues of agrochemicals. Organic farming practices such as the use of animal manure can however increase the risk of contamination by enteric pathogenic microorganisms and may consequently pose health risks. A number of scientific studies conducted in different countries have compared the microbiological quality of produce samples from organic and conventional production and results are contradictory. While some have reported greater microbial counts in fresh produce from organic production, other studies do not. This manuscript provides a brief review of the current knowledge and summarizes data on the occurrence of pathogenic microorganisms in vegetables from organic production.

Introduction

Fresh produce is a generalized term for a group of farm-produced crops, including fruits and vegetables. These foods are an important component of a healthy diet. Consumption of fresh produce is widely promoted by governmental health agencies since it supplies essential nutrients such as vitamins, minerals, dietary fiber and phytochemical compounds at a relatively low calorie density. Furthermore, the consumption of fruits and vegetables has been strongly associated with reduced chronic diseases, risk of heart disease and cancer.[1–3] Alternative cropping systems have been developed because of society's increasing concerns about the sustainability of conventional agriculture, intensive use of chemical products and their potential risk to human health and the environment.[4,5]

* *Corresponding author at*: Universidade de São Paulo, Faculdade de Ciências Farmacêuticas, Departamento de Alimentos e Nutrição Experimental, Av. Prof. Lineu Prestes, 580, 05508-000 São Paulo, SP, Brazil.
E-mail: danielemaffei@usp.br (D.F. Maffei).
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Organic agriculture has been on the rise and is attracting the attention of the food production sector in many parts of the world, since it revives eco-agricultural principles that are potentially more environmentally friendly and may provide products with few agrochemical residues.[6,7] Organic farming practices which use animal manure as fertilizer can increase the risk of contamination by enteric pathogenic microorganisms and, consequently, pose health risks becoming a major concern for consumers and governments. Furthermore, these foods are often consumed raw, increasing risk of infection if pathogens are present.[8]

Despite the growing demand for organic fresh produce and its health benefits, a number of foodborne disease outbreaks have been associated with the consumption of these foods.[8–14] However, the number of studies focusing on microbial safety of organically produced foods is low. This manuscript provides a brief review of the current knowledge and summarizes data on the risk of pathogenic microorganisms in vegetables from organic production.

Organic farming

The organic sector has expanded recently worldwide, due to policy support and a growing market demand for these products. Organic farming can be defined as an ecological production system that promotes and enhances biodiversity and biological cycle in soil, crop and livestock. It is based on minimal use of off-farm inputs and on management practices that restore, maintain and enhance ecological harmony.[15] The process of certification is also important in organic farming. Certification is intended to assure the consumers that a product marketed as organic was in fact produced according to organic production standards, which vary from country to country, based on their certifying bodies.[16]

Organic farming is regulated internationally by *Codex Alimentarius* Guidelines [established by the Food and Agricultural Organization of the United Nations (FAO) and the World Health Organization (WHO)] and by the International Federation of Organic Agriculture Movements (IFOAM) Basic Standards.[17] According to the IFOAM,[18] the principles of organic agriculture are: (i) health: organic agriculture should sustain and enhance the health of soil, plant, animal, human and planet as one and indivisible; (ii) ecology: organic agriculture should be based on living ecological systems and cycles, work with them, emulate them and help sustain them; (iii) fairness: organic agriculture should be built on relationships that ensure fairness with regard to the common environment and life opportunities and (iv) care: organic agriculture should be managed in a precautionary and responsible manner to protect the health and well-being of current and future generations and the environment.

Main sources of contamination of fresh produce by foodborne pathogens

Fresh produce can become contaminated with pathogenic microorganisms during pre-harvest (in the field) and post-harvest stages and this contamination can arise from environmental, animal or human sources. Pre-harvest sources include soil, irrigation water, inadequately composted or raw animal manure, dust, insects, presence of wild and domestic animals and human handling. Post-harvest sources include human handling, harvesting equipment, transport containers/vehicles, rinse water, improper storage and packaging.[8,11,19,20] Fig. 1 illustrates the main routes of fresh produce contamination in the field.

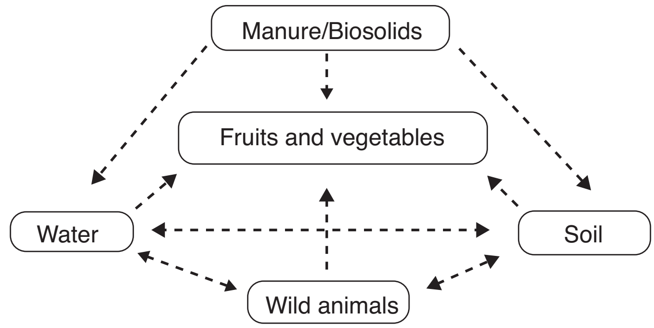

Fig. 1 – The sources and routes of contamination of fruits and vegetables.
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The soil is a habitat for many organisms, including human pathogens, which can contaminate plants through the seeds, roots or surface. Both conventional and organic produce can be fertilized with natural sources of nutrients such as animal manure and plant debris. Since animal manure is the main fertilizer type in organic farming, where no chemical treatment against bacteria is allowed, it gives rise to concern about the possible contamination of produce with microbial pathogens such as *Escherichia coli* O157:H7, *Salmonella* spp., and *Listeria monocytogenes*.[21,22] A key strategy used to reduce the concentration of enteric pathogens in manure is composting, the biological decomposition of organic matter by microorganisms under controlled conditions.[23] Compost can provide certain benefits to plants when applied to the soil. If composting is done incorrectly (i.e. for too short a time or at too low a temperature), the result can increase microbial proliferation and risk of pathogen contamination.[24,25]

The irrigation water can also be a source of contamination. The most common sources of water for irrigation include wells, rivers, reservoirs and lakes, all of which are susceptible to contamination by human pathogens. The presence of pathogenic microorganisms in irrigation water and their transfer to vegetables has been reported.[26–28] Moreover, vegetable cultivation in open areas allows the access of animals (birds, insects, rodents, domestic and wild animals), which can defecate in the fields and, therefore, be a source of contamination. Studies conducted by Islam et al.[29,30] showed that pathogens such as *S. Typhimurium* and *E. coli* O157:H7 can survive for a long period in soil (>150 days) and vegetables (>60 days) grown after experimental contamination using contaminated fertilizer (manure) or irrigation water.

Contaminated equipment, poor hygiene and improper post-harvest handling can compromise the microbial safety of a product. Proper storage conditions, including temperature, air circulation and relative humidity may be needed for pathogen control. The storage of fresh produce under refrigeration ($\leq$4 °C) is an important strategy to reduce the metabolic rate of the plant and prevent or limit the growth of pathogens.[31]

Pathogens isolated from or associated with organic produce

An increased number of foodborne disease outbreaks have been associated with the consumption of fresh produce recently.[8–14,32] According to a report by the Center for Science in the Public Interest (CSPI), fresh produce was the cause of most foodborne illnesses occurred in the U.S. between 2004 and 2013, including 193,754 illnesses from 9,625 outbreaks.[33] Studies have isolated pathogens including *Salmonella* spp., *L. monocytogenes* and pathogenic *E. coli* from fresh and fresh-cut vegetable samples in many countries.[34–49] Other studies have compared the microbiological quality of vegetables from organic and conventional production.[21,22,24,50–59] Ceuppens et al.[51] assessed the microbiological quality of lettuce production in Brazil and detected generic *E. coli* more frequently and at higher average concentrations in lettuce samples from organic farms (23.1% and 3.22 log CFU/g) vs. conventional farms (16.7% and 2.27 log CFU/g). Gomes Neto et al.[52] evaluated the microbiological quality of 180 iceberg lettuce samples from conventional ($n = 60$), organic ($n = 60$) and hydroponic ($n = 60$) cropping systems in Brazil and observed that samples from organic systems were the most contaminated both by bacteria and intestinal parasites, while the lowest contamination level was observed in hydroponically grown lettuce. Maffei et al.[24] analyzed 130 samples of different organic ($n = 65$) and conventional ($n = 65$) vegetable varieties (also in Brazil), and observed that some organic varieties had greater microbial counts with the highest incidence of generic *E. coli* in organic loose-leaf lettuce (90% of samples positive).

Mukherjee et al.[55] analyzed 476 organic and 129 conventional produce samples from farms in Minnesota, USA, and found greater prevalence of *E. coli* in organic vs. conventional samples. The largest prevalence of *E. coli* was in organic lettuce (22.4% of samples). Oliveira et al.[21] analyzed 72 lettuce samples of organic and conventional agriculture in Spain and found similar results, with a greater prevalence of *E. coli* in lettuce samples from organic (22.2%) than conventional (12.5%) agriculture. Wießner et al.[22] investigated the effect of different organic manures in comparison with mineral fertilizer on the risk of pathogen transfer in lettuce plants in Germany and observed that microbial counts tended to be slightly higher after organic fertilization, although the differences were not statistically significant ($p < 0.05$). Although these studies show that organic produce generally seems to be more contaminated than conventional produce, this effect was not seen in every study.

Bohaychuk et al.[50] analyzed 673 fresh produce samples collected from Alberta public and farmer's markets in Canada (including organic produce) and observed that the levels of *E. coli* in organically and conventionally grown produce was not significantly different ($p < 0.05$). Khalil and Gomaa[53] analyzed 380 samples of unpackaged whole conventional and 84 packaged whole organic leafy greens collected from retail markets in Alexandria, Egypt, and observed the mean total bacterial count for organic samples were statistically significantly less ($p < 0.05$) than those of the corresponding conventional samples and *E. coli* was detected in 100% of all leafy greens. Marine et al.[54] evaluated leafy greens from organic ($n = 178$) and conventional ($n = 191$) farms in the Mid-Atlantic Region of the United States. They observed that the farming system was not a significant factor for *E. coli*, aerobic mesophiles or *Salmonella*, but with (non-statistically significant) higher total coliform counts from organic farm samples.

Mukherjee et al.[56] conducted a 2-year study to evaluated 2029 preharvest produce samples (473 organic, 911 semiorganic, and 645 conventional) in Minnesota and Wisconsin, USA, and concluded that the microbiological quality of preharvest produce from the three types of farms was very similar. Phillips and Harrison[57] evaluated the microflora of organic ($n = 108$) and conventional ($n = 108$) spring mix samples obtained from a commercial California (USA) fresh-cut produce where manure is not used in the cultivation practices. The authors observed that the mean microbial population for conventional samples was not statistically different ($p > 0.05$) from the corresponding mean populations for organic samples. The mean population of each microbial group was significantly higher in unwashed vs. washed product. Ryu et al.[58] analyzed the microbiological quality of 11 types of environmentally friendly and conventionally grown vegetables sold at retail markets in Korea and did not observe significant difference ($p > 0.05$) in the overall microbiological quality among samples. Tango et al.[59] analyzed the microbiological quality of 354 Korean leafy vegetable samples (165 conventional and 189 organic) and their results did not support the hypothesis that organic produce poses a greater risk of pathogen contamination vs. conventional produce.

Only Ceuppens et al.,[51] Marine et al.,[54] Mukherjee et al.[55] and Tango et al.[59] detected pathogenic bacteria in the fresh produce samples analyzed above. Ceuppens et al.[51] isolated *Salmonella* from one organic lettuce sample. Marine et al.[54] isolated *Salmonella* from eight (2.16%) of 369 leafy greens [four organic (2.24%) and four conventional (2.09%) samples]. Mukherjee et al.[55] isolated *Salmonella* from one organic lettuce and one organic green pepper sample. Tango et al.[59] reported positive results for *Bacillus cereus* ($n = 17$), *Staphylococcus aureus* ($n = 13$) and *L. monocytogenes* ($n = 10$) in 63 organic samples, vs. 3, 8 and 6, respectively for conventional samples. These authors also detected *E. coli* O157:H7 in 1 out of 55 conventional samples.

Some scientific studies have been conducted to determine the presence of pathogenic bacteria exclusively in organic produce.[60–67] Among these, only Chang et al.,[61] Loncarevic et al.,[62] McMahon and Wilson,[64] Nguz et al.[65] and Rodrigues et al.[66] detected the presence of pathogenic bacteria. Chang et al.[61] reported the presence of *E. coli* O157:H7 in four out of 210 organic vegetables collected in supermarket and groceries in Selangor, Malaysia. Loncarevic et al.[62] isolated *L. monocytogenes* serogroups 1 and 4 from two of 179 organically grown leaf lettuce samples in Norway. McMahon and Wilson[64] investigated the occurrence of selected enteric pathogens and *Aeromonas* species in organic vegetables in Northern Ireland. They found *Aeromonas* spp only (34% of the samples)

Table 1 – Incidence of pathogenic bacteria on organic vegetables.

Country	Pathogen	Number of samples		Reference
		Total *n*	Positive *n* (%)	
Brazil	*Salmonella* spp.	75	1 (1.33)	Ceuppens et al.[51]
Brazil	*Salmonella* spp.	36	1 (2.77)	Rodrigues et al.[66]
Korea	*B. cereus*	63	17 (26.9)	Tango et al.[59]
	L. monocytogenes	63	10 (15.8)	
	S. aureus	63	13 (20.6)	
Malaysia	*E. coli* O157:H7	210	4 (1.90)	Chang et al.[61]
Northern Ireland	*Aeromonas* spp.	86	29 (34.0)	McMahon and Wilson[64]
Norway	*L. monocytogenes*	179	2 (1.11)	Loncarevic et al.[62]
USA	*Salmonella* spp.	178	4 (2.24)	Marine et al.[54]
USA	*Salmonella* spp.	476	2 (0.42)	Mukherjee et al.[55]
Zambia	*L. monocytogenes*	80	16 (20.0)	Nguz et al.[65]
	Salmonella spp.	160	37 (23.1)	
	S. aureus	80	54 (80.0)	

Data based on studies that presented positive results (presence of pathogens).

including *A. schubertii* (21%), *A. hydrophila* (5.8%), *A. trota* (5.8%), *A. caviae* (3.5%) and *A. veronii* biovar *veronii* (2.3%). These *Aeromonas* species were previously reported to be potentially pathogenic and responsible for gastrointestinal infections in humans.[68,69] Nguz et al.[65] assessed the microbiological quality of fresh-cut organic vegetables (washed with chlorine solution at 150 $\mu g\,mL^{-1}$) produced in Zambia and detected the presence of *L. monocytogenes*, *Salmonella* and *S. aureus* in 20%, 23.1% and 80.0% of samples, respectively. Rodrigues et al.[66] isolated *Salmonella* from only one of 36 Brazilian organic lettuce samples.

The incidence of pathogens in organic vegetables varies according to the study (Table 1). *Salmonella* spp prevalence varies between 0.4 and 23.1%, *L. monocytogenes* between 1.1 and 20% and *S. aureus* between 20.6 and 80%. *B. cereus* prevalence can be as high as 26.9% and as high as 34% for *Aeromonas* spp. *E. coli* O157:H7 prevalence can be as high as 1.9%. Since fresh produce is often consumed raw even low prevalence rates may mean increased risk of foodborne disease from these foods.

Information on foodborne outbreaks from organic produce is limited. However, some cases involving Shiga toxin-producing *E. coli* (STEC) and organic produce have been reported in the past few years. An outbreak involving STEC O157:H7 linked to organic spinach and spring mix blend occurred in the U.S. in 2012 and affected 33 persons from five states: 46% were hospitalized and two developed hemolytic uremic syndrome (HUS).[70] STEC O104:H4 was responsible for an outbreak linked to organic raw sprouts in Germany, with thousands of infections.[71,72]

Control measures

Current strategies to control microbial contamination during the production of fresh produce (regardless of cultivation method) are based on the implementation of Good Agricultural Practices (GAP). A GAP framework considers the implementation of best practices regarding worker's health and hygiene, soil and water quality, sewage treatment, wildlife and livestock management, manure and biosolids management, field sanitation and hygiene, and harvest and transportation.[20] The implementation of food safety management tools such as Good Hygienic Practices (GHP) and Hazard Analysis and Critical Control Points (HACCP) at postharvest steps also helps to reduce, eliminate or prevent the occurrence of hazards. The washing step before consumption is also important as it may reduce microbial load on vegetables surfaces. Washing with sanitizer may also reduce contamination and prevent cross-contamination by pathogenic microorganisms.[73–76]

Concluding remarks

Data summarized above from a variety of studies focusing on different crops and different countries highlight the potential risks of pathogenic microorganisms in fresh produce, including organic products. Although a number of studies have indicated that organic produce may pose a greater risk than conventional grown produce, this trend is not universal across all studies. Further efforts are needed to understand and control the disease risk associated with organic produce from harvest through consumption.

Conflicts of interest

The authors declare no conflicts of interest.

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REFERENCES

1. Dias JS. Nutritional quality and health benefits of vegetables: a review. *Food Nutr Sci*. 2012;3:1354–1374.
2. Liu RH. Health-promoting components of fruits and vegetables in the diet. *Adv Nutr*. 2013;4:384–392.
3. Slavin JL, Lloyd B. Health benefits of fruits and vegetables. *Adv Nutr*. 2012;3:506–516.
4. Bettiol W, Ghini R, Galvão JAH, Siloto RC. Organic and conventional tomato cropping systems. *Sci Agric*. 2004;61:253–259.
5. Crowder DW, Reganold JP. Financial competitiveness of organic agriculture on a global scale. *Proc Natl Acad Sci U S A*. 2015;112:7611–7616.
6. Azadi H, Schoonbeek S, Mahmoudi H, Derudder B, De Maeyer P, Witloxa F. Organic agriculture and sustainable food production system: main potentials. *Agric Ecosyst Environ*. 2011;144:92–94.
7. Willer H, Lernoud J, eds. *The World of Organic Agriculture. Statistics and Emerging Trends*. Bonn: Research Institute of Organic Agriculture (FiBL), Frick, and IFOAM – Organics International; 2016.
8. Berger CN, Sodha SV, Shaw RK, et al. Fresh fruit and vegetables as vehicles for the transmission of human pathogens. *Environ Microbiol*. 2010;12:2385–2397.
9. Callejón RM, Rodríguez-Naranjo MI, Ubeda C, Hornedo-Ortega R, Garcia-Parrilla MC, Troncoso AM. Reported foodborne outbreaks due to fresh produce in the United States and European Union: trends and causes. *Foodborne Pathog Dis*. 2015;12:32–38.
10. Doyle MP, Erickson MC. Summer meeting 2007 – the problems with fresh produce: an overview. *J Appl Microbiol*. 2008;105:317–330.
11. Harris LJ, Farber JN, Beuchat LR, et al. Outbreaks associated with fresh produce: incidence, growth, and survival of pathogens in fresh and fresh-cut produce. *Compr Rev Food Sci Food Saf*. 2003;2:78–141.
12. Jung Y, Jang H, Matthews KR. Effect of the food production chain from farm practices to vegetable processing on outbreak incidence. *Microb Biotechnol*. 2014;7:517–527.
13. Lynch MF, Tauxe RV, Hedberg CW. The growing burden of foodborne outbreaks due to contaminated fresh produce: risks and opportunities. *Epidemiol Infect*. 2009;137:307–315.
14. Sivapalasingam S, Friedman CR, Cohen L, Tauxe RV. Fresh produce: a growing cause of outbreaks of foodborne illness in the United States, 1973 through 1997. *J Food Prot*. 2004;67:2342–2353.
15. Winter CK, Davis SF. Organic foods. *J Food Sci*. 2006;71:117–124.
16. Vijayan G. *Organic Food Certification and Marketing Strategies*. Agrihortico; 2014.
17. Tuomisto HL, Hodge ID, Riordan P, Macdonald DW. Does organic farming reduce environmental impacts? A meta-analysis of European research. *J Environ Manag*. 2012;112:309–320.
18. International Federation of Organic Agriculture Movements (IFOAM). *The Principles of Organic Agriculture*. Bonn, Germany: Preamble. IFOAM; 2005. Available at http://www.ifoam.org/sites/default/files/ifoam_poa.pdf Accessed 06.09.16.
19. Beuchat LR. Ecological factors influencing survival and growth of human pathogens on raw fruits and vegetables. *Microbes Infect*. 2002;4:413–423.
20. Sant'Ana AS, Silva FFP, Maffei DF, Franco BDGM. Fruits and vegetables: introduction. In: Batt CA, Tortorello ML, eds. *Encyclopedia of Food Microbiology*. 2nd ed. Amsterdam: Academic Press; 2014:972–982.
21. Oliveira M, Usall J, Viñas I, Anguera M, Gatius F, Abadias M. Microbiological quality of fresh lettuce from organic and conventional production. *Food Microbiol*. 2010;27:679–684.
22. Wießner S, Thiel B, Krämer J, Köpke U. Hygienic quality of head lettuce: effects of organic and mineral fertilizers. *Food Control*. 2009;20:881–886.
23. Moreno J, López MJ, Vargas-García MC, Suárez-Estrella F. Recent advances in microbial aspects of compost production and use. *Acta Hort (ISHS)*. 2013;1013:443–457.
24. Maffei DF, Silveira NFA, Catanozi MPLM. Microbiological quality of organic and conventional vegetables sold in Brazil. *Food Control*. 2013;29:226–230.
25. Suárez-Estrella F, Vargas-García MC, Elorrieta MA, López MJ, Moreno J. Temperature effect on *Fusarium oxysporum* f.sp. *melonis* survival during horticultural waste composting. *J Appl Microbiol*. 2003;94:475–482.
26. Chigor VN, Umoh VJ, Smith SI. Occurrence of *Escherichia coli* O157 in a river used for fresh produce irrigation in Nigeria. *Afr J Biotechnol*. 2010;9:178–182.
27. Ijabadeniyi OA, Debusho LK, Vanderlinde M, Buys EM. Irrigation water as a potential preharvest source of bacterial contamination of vegetables. *J Food Saf*. 2011;31:452–461.
28. Okafo CN, Umoh VJ, Galadima M. Occurrence of pathogens on vegetables harvested from soils irrigated with contaminated streams. *Sci Total Environ*. 2003;311:49–56.
29. Islam M, Doyle MP, Phatak SC, Millner P, Jiang X. Persistence of enterohemorrhagic *Escherichia coli* O157:H7 in soil and on leaf lettuce and parsley grown in fields treated with contaminated manure composts or irrigation water. *J Food Prot*. 2004;67:1365–1370.
30. Islam M, Morgan J, Doyle MP, Phatak SC, Millner P, Jiang X. Persistence of *Salmonella enterica* serovar Typhimurium on lettuce and parsley and in soils on which they were grown in fields treated with contaminated manure composts or irrigation water. *Foodborne Pathog Dis*. 2004;1:27–35.
31. Matthews KR. Sources of enteric pathogen contamination of fruits and vegetables: future directions of research. *Stewart Postharvest Rev*. 2013;9:1–5.
32. Sagoo SK, Little CL, Ward L, Gillespie IA, Mitchell RT. Microbiological study of ready-to-eat salad vegetables from retail establishments uncovers a national outbreak of salmonellosis. *J Food Prot*. 2003;66:403–409.
33. Center for Science in the Public Interest. *Outbreak Alert! 2015: A Review of Foodborne Illness in the U.S. from 2004–2013*; 2015. Available at: https://cspinet.org/reports/outbreak-alert-2015.pdf. Accessed 25 August 2016.
34. Abadias M, Usall J, Anguera M, Solsona C, Viñas I. Microbiological quality of fresh, minimally-processed fruit and vegetables, and sprouts from retail establishments. *Int J Food Microbiol*. 2008;123:121–129.
35. Arthur L, Jones S, Fabri M, Odumeru J. Microbial survey of selected Ontario-grown fresh fruits and vegetables. *J Food Prot*. 2007;70:2864–2867.
36. De Léon HB, Gómez-Aldapa CA, Rangel-Vargas E, Vázquez-Barrios E, Castro-Rosas J. Frequency of indicator bacteria, *Salmonella* and diarrhoeagenic *Escherichia coli* pathotypes on ready-to-eat cooked vegetable salads from Mexican restaurants. *Lett Appl Microbiol*. 2013;56:414–420.
37. Froder H, Martins CG, Souza KLO, Landgraf M, Franco BDGM, Destro MT. Minimally processed vegetable salads: microbial quality evaluation. *J Food Prot*. 2007;70:1277–1280.
38. Giusti M, Aurigemma C, Marinelli L, et al. The evaluation of the microbial safety of fresh ready-to-eat vegetables produced by different technologies in Italy. *J Appl Microbiol*. 2010;109:996–1006.
39. Jeddi MZ, Yunesian M, Gorji ME, Noori N, Pourmand MR, Khaniki GRJ. Microbial evaluation of fresh,

minimally-processed vegetables and bagged sprouts from chain supermarkets. *J Health Popul Nutr*. 2014;32:391–399.

40. Kovacevic M, Burazin J, Pavlovic H, Kopjar M, Pilizota V. Prevalence and level of *Listeria monocytogenes* and other *Listeria* sp. in ready-to-eat minimally processed and refrigerated vegetables. *World J Microb Biotechnol*. 2013;29:707–712.
41. Maistro LC, Miya NTN, Sant'Ana AS, Pereira JL. Microbiological quality and safety of minimally processed vegetables marketed in Campinas, SP – Brazil, as assessed by traditional and alternative methods. *Food Control*. 2012;28:258–264.
42. Mora A, León SL, Blanco M, et al. Phage types, virulence genes and PFGE profiles of Shiga toxin-producing *Escherichia coli* O157:H7 isolated from raw beef, soft cheese and vegetables in Lima (Peru). *Int J Food Microbiol*. 2007;114:204–210.
43. Moreno Y, Sánchez-Contreras J, Montes RM, García-Hernández J, Ballesteros L, Ferrús MA. Detection and enumeration of viable *Listeria monocytogenes* cells from ready-to-eat and processed vegetable foods by culture and DVC-FISH. *Food Control*. 2012;27:374–379.
44. Oliveira MA, Souza VM, Bergamini AMM, Martinis ECP. Microbiological quality of ready-to-eat minimally processed vegetables consumed in Brazil. *Food Control*. 2011;22:1400–1403.
45. Quiroz-Santiago C, Rodas-Suárez OR, Vázquez QCR, Fernández FJ, Quiñones-Ramírez EI, Vázquez-Salinas C. Prevalence of *Salmonella* in vegetables from Mexico. *J Food Prot*. 2009;72:1279–1282.
46. Rúgeles LC, Bai J, Martínez AJ, Vanegas MC, Gómez-Duarte OG. Molecular characterization of diarrheagenic *Escherichia coli* strains from stools samples and food products in Colombia. *Int J Food Microbiol*. 2010;138:282–286.
47. Sant'Ana AS, Igarashi MC, Landgraf M, Destro MT, Franco BDGM. Prevalence, populations and pheno- and genotypic characteristics of *Listeria monocytogenes* isolated from ready-to-eat vegetables marketed in São Paulo, Brazil. *Int J Food Microbiol*. 2012;155:1–9.
48. Sant'Ana AS, Landgraf M, Destro MT, Franco BDGM. Prevalence and counts of *Salmonella* spp. in minimally processed vegetables in São Paulo, Brazil. *Food Microbiol*. 2011;28:1235–1237.
49. Seo Y, Jang J, Moon K. Microbial evaluation of minimally processed vegetables and sprouts produced in Seoul, Korea. *Food Sci Biotechnol*. 2010;19:1283–1288.
50. Bohaychuk VM, Bradbury RW, Dimock R, et al. A microbiological survey of selected Alberta-grown fresh produce from farmers' markets in Alberta, Canada. *J Food Prot*. 2009;72:415–420.
51. Ceuppens S, Hessel CT, Rodrigues RQ, Bartz S, Tondo EC, Uyttendaelea M. Microbiological quality and safety assessment of lettuce production in Brazil. *Int J Food Microbiol*. 2014;181:67–76.
52. Gomes Neto NJ, Pessoa RML, Queiroga IMBN, et al. Bacterial counts and the occurrence of parasites in lettuce (*Lactuca sativa*) from different cropping systems in Brazil. *Food Control*. 2012;28:47–51.
53. Khalil R, Gomaa M. Evaluation of the microbiological quality of conventional and organic leafy greens at the time of purchase from retail markets in Alexandria, Egypt. *Pol J Microbiol*. 2014;63:237–243.
54. Marine SC, Pagadala S, Wang F, et al. The growing season, but not the farming system, is a food safety risk determinant for leafy greens in the Mid-Atlantic region of the United States. *Appl Environ Microb*. 2015;8:2395–2407.
55. Mukherjee A, Speh D, Dyck E, Diez-Gonzales F. Preharvest evaluation of coliforms, *Escherichia coli*, *Salmonella*, and *Escherichia coli* O157:H7 in organic and conventional produce grown by Minnesota farmers. *J Food Prot*. 2004;67: 894–900.
56. Mukherjee A, Speh D, Jones AT, Buesing KM, Diez-Gonzalez F. Longitudinal microbiological survey of fresh produce grown by farmers in the upper midwest. *J Food Prot*. 2006;69:1928–1936.
57. Phillips CA, Harrison MA. Comparison of the microflora on organically and conventionally grown spring mix from a California processor. *J Food Prot*. 2005;68:1143–1146.
58. Ry JH, Kim M, Kim EG, Beuchat LR, Kim H. Comparison of the microbiological quality of environmentally friendly and conventionally grown vegetables sold at retail markets in Korea. *J Food Sci*. 2014;79:M1739–M1744.
59. Tango CN, Choi NJ, Chung MS, Oh DH. Bacteriological quality of vegetables from organic and conventional production in different areas of Korea. *J Food Prot*. 2014;77: 1411–1417.
60. Batalha EY [M. Sc. Dissertation] *Escherichia coli produtora de toxina de Shiga em vegetais orgânicos cultivados na região metropolitana de SP*. São Paulo, Brasil: Faculdade de Ciências Farmacêuticas. USP, São Paulo; 2015, 64 pp.
61. Chang WS, Afsah-Hejri L, Rukayadi Y, et al. Quantification of *Escherichia coli* O157:H7 in organic vegetables and chickens. *Int Food Res J*. 2013;20:1023–1029.
62. Loncarevic S, Johannessen GS, Rorvik LM. Bacteriological quality of organically grown leaf lettuce in Norway. *Lett Appl Microbiol*. 2005;41:186–189.
63. Machado DC, Maia CM, Carvalho ID, da Silva NF, Andre M.C.D.P.B., Serafini AB. Microbiological quality of organic vegetables produced in soil treated with different types of manure and mineral fertilizer. *Braz J Microbiol*. 2006;37:538–544.
64. McMahon MAS, Wilson IG. The occurrence of enteric pathogens and *Aeromonas* species in organic vegetables. *Int J Food Microbiol*. 2001;70:155–162.
65. Nguz K, Shindano J, Samapundo S, Huyghebaert A. Microbiological evaluation of fresh cut organic vegetables produced in Zambia. *Food Control*. 2005;16:623–628.
66. Rodrigues RQ, Loiko MR, de Paula CMD, et al. Microbiological contamination linked to implementation of good agricultural practices in the production of organic lettuce in Southern Brazil. *Food Control*. 2014;42:152–164.
67. Sagoo SK, Little CL, Mitchell RT. The microbiological examination of ready-to-eat organic vegetables from retail establishments in the United Kingdom. *Lett Appl Microbiol*. 2001;33:434–439.
68. Igbinosa IH, Igumbor EU, Aghdasi F, Tom M, Okoh A. Emerging *Aeromonas* species infections and their significance in public health. *Sci World J*. 2012:1–13.
69. Merino S, Rubines X, Knichel S, Tomás JM. Emerging pathogens: *Aeromonas* spp. *Int J Food Microbiol*. 1995;28:157–168.
70. Centers for Disease Control and Prevention. *Multistate Outbreak of Shiga Toxin-Producing Escherichia coli O157:H7 Infections Linked to Organic Spinach and Spring Mix Blend (Final Update)*; 2012. Available at http://www.cdc.gov/ecoli/2012/O157H7-11-12/index.html Accessed 25.08.16.
71. Frank C, Werber D, Cramer JP, et al., HUS Investigation Team. Epidemic profile of shiga-toxin-producing *Escherichia coli* O104:H4 Outbreak in Germany. *N Engl J Med*. 2011;365:1771–1780.
72. King LA, Nogareda F, Weill FX, et al. Outbreak of shiga toxin-producing *Escherichia coli* O104:H4 associated with organic fenugreek sprouts, France, June 2011. *Clin Infect Dis*. 2012;54:1588–1594.
73. López-Gálvez F, Allende A, Selma MV, Gil MI. Prevention of *Escherichia coli* cross contamination by different commercial

sanitizers during washing of fresh-cut lettuce. *Int J Food Microbiol*. 2009;133:167–171.
74. Maffei DF, Sant'Ana AS, Monteiro G, Schaffner DW, Franco BDGM. Assessing the effect of sodium dichloroisocyanurate concentration on transfer of *Salmonella enterica* serotype Typhimurium in wash water for production of minimally processed iceberg lettuce (*Lactuca sativa* L). *Lett Appl Microbiol*. 2016;62:444–451.
75. Tomás-Callejas A, López-Gálvez F, Sbodio A, Artés F, Artés-Hernández F, Suslow TV. Chlorine dioxide and chlorine effectiveness to prevent *Escherichia coli* O157:H7 and *Salmonella* cross contamination on fresh-cut Red Chard. *Food Control*. 2012;23:325–332.
76. Zhang G, Ma L, Phelan VH, Doyle MP. Efficacy of antimicrobial agents in lettuce leaf processing water for control of *Escherichia coli* O157:H7. *J Food Prot*. 2009;72:1392–1397.