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Microbial dynamics of indicator microorganisms on fresh tomatoes in the supply chain from Mexico to the USA

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ABSTRACT

Quality and safety of fresh produce are important to public health and maintaining commerce between Mexico and USA. While preventive practices can reduce risks of contamination and are generally successful, the variable environment of the supply chain of fresh produce can be suitable for introduction or proliferation of pathogenic microorganisms. As routine surveillance of these pathogens is not practical, indicator microorganisms are used to assess the sanitary conditions of production and handling environments. An opportunity exists to use indicators on fresh produce to measure how handling and transport from field to market may affect microbial populations that contribute to their quality or safety. The objective was to quantify indicator microorganisms on tomatoes sampled along the supply chain during the harvest year, in order to observe the levels and changes of populations at different locations. Roma tomatoes ($n = 475$) were taken from the same lots ($n = 28$) at four locations of the postharvest supply chain over five months: at arrival to and departure from the packinghouse in México, at the distribution center in Texas, and at retail in USA. Samples were analyzed individually for four microbial populations: aerobic plate count (APC), total coliforms (TC), generic *Escherichia coli*, and yeasts and molds (YM). APC population differed ($p < 0.05$) from 1.9 ± 1.1, 1.7 ± 1.1, 2.3 ± 1.1 and 3.5 ± 1.4 log CFU/g at postharvest, packing, distribution center and supermarket, respectively. TC populations were <1 log CFU/g at postharvest, increased at packing (0.7 ± 1.0 log CFU/g), decreased in distribution (0.4 ± 0.8 log CFU/g) and increased in supermarkets (1.4 ± 1.5 log CFU/g). Generic *E. coli* was not identified from coliform populations in this supply chain. YM populations remained <1 log CFU/g, with the exception of 1.1 ± 1.3 log CFU/g at supermarkets and tomatoes were not visibly spoiled. The levels reported from this pilot study demonstrated the dynamics within populations as influenced by time and conditions in one supply chain during a harvest year, while the large variances in some locations indicate opportunities for improvement. Overall, packinghouse and supermarket locations were identified as crucial points to control microbial safety risks.

1. Introduction

The supply chain of fresh produce from Mexico to the United States is important to the health and well-being of consumers and to the industry's commerce. A wide variety of fresh fruit and vegetable products traverse this border each year, totaling 13 billion pounds worth over 6.2 billion US dollars. While there is domestic production of fresh tomatoes, imports account for about half of US consumption and originate mainly from Mexico (85%) and Canada (13%). The imported fresh tomato category includes the following varieties: cherry (2%), grape (4%), round (16%), Roma (37%), and hothouse/greenhouse (41%). Mexico holds 71% market share of imported hothouse tomatoes and 99% market share of imported Roma tomatoes (Wells, 2015; 2016). Regardless of their origin, fresh tomatoes are generally hand-picked and consumed raw, making both the quality and safety of these products essential for maintaining this industry. As such, the supply chain is specifically designed to both preserve and monitor attributes of the product and its production environment. Programs such as Good Agricultural Practices (GAPs) are in place to reduce the risk of product degradation and contamination in production, harvest and handling environments. Third party auditing groups serve to verify the legitimacy of such practices and records within each operation. Postharvest handling practices specific to quality include culling damaged products after harvest, washes, sanitizer treatment, storage and transportation under controlled atmosphere conditions, and visual inspection upon receipt of the product at distribution and retail centers.

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Still, there exist processes and conditions suitable for introduction, survival or growth of microorganisms that can affect produce safety or quality as it travels from field to the point of sale (Allen et al., 2005; Beuchat and Mann, 2008; Brar and Danyluk, 2013; Buchholz et al., 2012a,b; Jensen et al., 2013; McCollum et al., 2013; Perez-Rodriguez et al., 2014; Sreedharan et al., 2014; Wang and Ryser, 2014b). Fifteen multi-state outbreaks (1959 illnesses) of *Salmonella* in the USA between1990–2010 were associated with round (69%), Roma (23%), and grape (8%) tomatoes. Although epidemiological studies linked cases to consumption of domestically produced tomatoes in restaurants, traceback investigations into contamination sources were often complicated by the web of grower, packers, distributers and retailers that handled the product (Bennett et al., 2015). Pathogen surveillance in the final product is not necessarily a practical or successful measure for a farm, packinghouse or distribution center to use in order to assess the quality or safety of a fresh produce item. Depending on the microorganism, prevalence may be low, results can take up a considerable amount of the product's shelf life, the entire lot must be held and possibly removed from commerce, and the results may not indicate the source of the problem.

Instead, indicator microorganisms, such as *Enterobacteriaceae*, coliforms and generic *Escherichia coli*, may be used in assessments of the overall quality of a product and the hygienic conditions present in its production and handling environments (Kornacki, 2001). In this study, indicator organisms were used for understanding the potential influences of the supply chain on microbial populations on fresh produce. Roma tomatoes produced in Mexico and exported to several retail markets in different states of the USA were followed to study these hypothesized population dynamics, with the objective of quantifying the magnitude of changes due to conditions along the supply chain during one harvest year.

2. Materials and methods

2.1. The supply chain and sampling locations

Refer to Fig. 1 for the overall sampling design. Roma tomatoes were produced on a farm located in the state of Nuevo León, México. This farm utilized protected agriculture systems of greenhouses and shade houses on forty-two acres, which corresponded to different lot codes of the final products. Drip fertigation of plants drew water from deep wells on-site. Tomatoes were hand-harvested, with stems removed, into plastic containers. Tomatoes were transported to a packinghouse located within the same farm, where they were spray-washed with chlorinated water and brush rollers. 150 ppm total chlorine was measured and maintained in the wash water every hour using test strips (Diken International, Monterrey, Mexico) and completely changed every 4 h or 24 pallets, whichever came first. After washing and sanitizing, tomatoes were forced-air dried on rollers, conveyed through sorting and hand-packed into boxes. The first sampling location was upon arrival to the packinghouse after harvesting, where tomato samples were taken with gloved hands from their plastic harvesting containers (referred to as "harvest"). The second sampling location was after the washing and sorting steps and immediately prior to boxing, where tomato samples were taken by the workers ("packing" or "packinghouse"). The same day, palletized boxes were loaded onto refrigerated trailer trucks for transportation to and storage in a distribution center in southern Texas, USA. The third sampling location ("distribution") was palletized boxes in the cold storage room (9–10 °C) of the distribution center, with gloved hands after several days of storage and prior to shipment to clients. Tomato boxes were sold based on size and color to retail supermarkets in Texas, North Carolina, Minnesota, and Michigan, USA. The final sampling location ("supermarket") was the point of sale in supermarkets or in one occasion from the supermarket storage room, again with gloved hands. In all instances, the same lot codes designated by the farm and displayed on each box were followed through the supply chain for a total of 28 different lots through distribution and 11 of those lots through retail, over one production season.

Tomato samples (4–10 fruits) were taken individually in Ziploc bags from each lot code depending on the sampling location and were maintained on ice until individual analysis (within 48 h). A total of 475 tomatoes were taken throughout the supply chain: 130 tomatoes at postharvest, 130 at packing, 144 at the distribution center, and 71 from five different supermarkets. Difficulty in traceability of lots explains the different number of samples at each location along the supply chain.

Control samples (n = 30 fruits) were taken from the harvest location and maintained at 10–12 °C, 90% relative humidity (RH) for up to ten days. Similarly, additional control samples (n = 30 fruits) were taken immediately after the chlorinated wash and maintained 10–12 °C, 90% RH for up to ten days. These tomatoes did not travel the supply chain, but were maintained in storage conditions typical for maximizing postharvest quality during distribution and storage (Suslow and Cantwell, 1997) and analyzed for microbial indicators every other day.

2.2. Microbiological analyses

Tomato samples were analyzed at the Autonomous University of Nuevo León (San Nicolas, NL México) or Cornell University (Ithaca, NY USA) following the same protocol: Into the bag containing the tomato sample (83 ± 20 g), an equal volume to weight ratio of 0.1% peptone water was added and the tomato surface was washed by gentle rubbing for 1 min. All enumeration methods followed the pour plate technique using 1 ml, 0.1 ml and 0.01 ml of the rinse water. For aerobic plate count, serial dilutions were plated in duplicate using Standard Methods

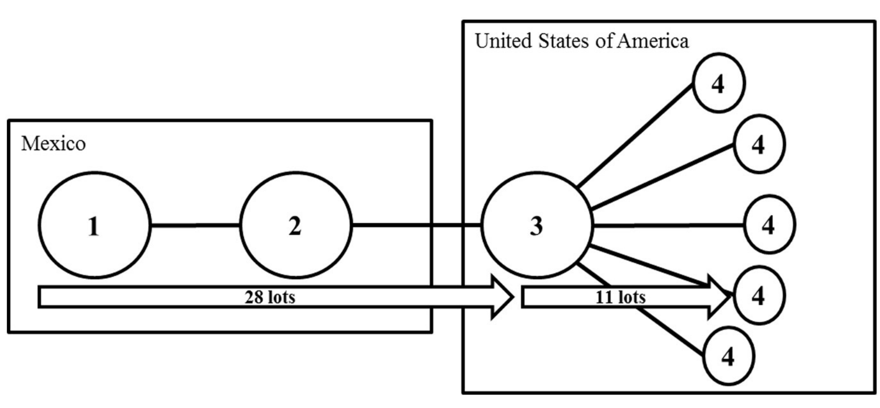

Fig. 1. Schematic of the supply chain sampling from Nuevo Leon, Mexico to several supermarkets in the US. The following locations were sampling points for a total of 28 lots of tomatoes: (1) Arrival to packinghouse (n = 130 tomatoes); (2) End of the packing line (n = 130); (3) Distribution center storage room (n = 144). Of those 28 lots, only 11 lots were recovered in the final location: (4) Retail supermarkets in the United States (n = 71).