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Assessment of foodborne pathogen presence in the peach supply chain and its potential risk to the end consumer

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ABSTRACT

Peaches are popular, nutritious and widely consumed. Being a tree crop, it is considered a low risk fruit, with no direct water contact, and no previous foodborne disease outbreaks associated with its consumption. However, in 2014 the pioneer association between stone fruit and a foodborne illness was reported, linking *Listeria monocytogenes* to stone fruit. This highlights the need for better understanding of risk associated with contaminated fresh stone fruit, in order to implement adequate preventative measures. No information is available on the presence of foodborne pathogens on peaches in the supply chain. A case study approach was therefore followed to assess foodborne pathogen presence on the farm, focusing on the impact of irrigation water, facility sanitation and hygiene by collecting various fruit and environmental samples (n = 428). This study demonstrates the effectiveness of integrating basic microbial testing with safety management and risk assessment tools that can be collectively used to improve the food safety management system. No *Salmonella* Typhimurium was detected from samples, however, *Escherichia coli* O157:H7, *Listeria* spp. and *Staphylococcus aureus* were detected on fruit and environmental samples. Despite the GlobalG.A.P. certification status of the farm, livestock frequented water sources which lead to *E. coli* O157:H7 contamination. This conclusion was based on positive detection of foodborne pathogens from the water sources and subsequent removal of livestock which resulted in a definite decrease in pathogen detection. A number of *E. coli* O157:H7 and *S. aureus* were detected during the second year of monitoring from environmental samples and it was observed that the personal hygiene and facility sanitation was not adequately enforced. Based on feedback given to the farmer, enforcement was improved and a definite decrease in foodborne pathogens was observed in the following sampling cycle. Areas of risk that were still identified following the fourth year of monitoring included the water source used for irrigation and poor sanitation in the production and processing facilities. Limited foodborne pathogen prevalence on peaches over the full study period as well as the extended export supply chain at controlled temperatures resulted in low-to-medium calculated consumer risk. The correct and meticulous implementation of integrated and holistic pre- and post-harvest food safety management systems is therefore essential to prevent produce contamination, reduce the consumer risk and therefore ensure overall product safety.

1. Introduction

Escherichia coli O157:H7, *Listeria monocytogenes* and *Salmonella* spp. are well described foodborne pathogens, having been associated with several disease outbreaks on fresh produce. Stone fruit are not traditionally considered a high-risk product due to production practices. However, in recent years' commodities previously not associated with foodborne disease outbreaks are becoming implicated, as was the case with the recent illnesses associated with *L. monocytogenes* on stone fruit (Jackson et al., 2015) and caramel apples [Centre for Disease Control and Prevention (CDC), 2015]. Due to extensive global distribution of fresh produce, outbreaks are not confined to the country of origin, as was the case in the June 2011 *E. coli* O104:H4 outbreak associated with contaminated sprouts, imported from Egypt, which affected 16 countries including 14 countries in the European Union (EU) as well as the United States of America (USA) and Canada [European Food Safety Authority (EFSA) (2011); World Health Organisation

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Abbreviations

CDC	Centre for Disease Control and Prevention
EFSA	European Food Safety Authority
EU	European Union
HSMS	Horticultural safety management system
RRR	Risk Ranger ranking
SA	South Africa
UK	United Kingdom
USA	United States of America
WHO	World Health Organisation

(WHO), 2011]. Food safety assurance is therefore of global importance.

Peaches are packed and consumed raw without any decontamination, it is therefore essential to prevent contamination. Preharvest contamination can occur through contact with contaminated soil, irrigation water and improperly composted manure (Beuchat, 2002). The presence of animal farming in fields adjacent to cultivation areas (Gruszynski et al., 2014; Kilonzo et al., 2013) or cultivation in fields which are historically used for animal rearing (Tauxe et al., 1997) could lead to the spread of persistent foodborne pathogens. Postharvest contamination usually occurs through contact with contaminated harvesting equipment, handlers and contact surfaces (Beuchat, 2002; Warriner, Huber, Namvar, Fan, & Dunfield, 2009).

Food safety standards and systems that target zero microbial contamination have been developed specifically for the food processing industries. However, the general philosophy is that zero tolerance is not realistic in a preharvest environment. Since microbiological analysis of food is time consuming, the International Commission on Microbiological Specification for Foods stated that "*Good Agricultural Practices and acceptable hygienic farming practices are more important than microbiological testing of food samples before selling*" (Food Science Australia, 2000). Good agricultural practices have been standardly adopted by the fresh produce industry and GlobalG.A.P. has become a global benchmark for exported produce. The use and implementation of effective food safety management systems should therefore provide additional food safety confidence.

The overall aim of this study was to determine 1) hazard presence, 2) on-farm risk areas and 3) end-consumers risk. The presence of *E. coli* O157:H7, *Listeria* spp., *Salmonella* Typhimurium and *Staphylococcus aureus* in water, on pre- and post-harvest fruit and pre- and post-harvest contact surfaces was determined. The overall risk and risk areas as well as control were assessed using a self-diagnostic tool (Kirezieva, Luning, Jacxsens, & Uyttendaele, 2015; Kirezieva et al., 2013) and an easy-to-use semi-quantitative risk assessment tool (Ross & Sumner, 2002).

2. Experimental

2.1. Study site and sampling strategy

An integrated commercial export farm managed according to industry guidelines and GlobalG.A.P. certified in the Limpopo Province, South Africa (SA) was selected as the site. This farm cultivated peaches, maize and citrus, in addition, the farm engaged in compost production, game and cattle livestock farming. Peaches (*Prunus persica* L.) were grown in uncovered fields, drip irrigated and conventional pesticide application with water sourced from on-farm collection dams filled with water from the Lephalala River. The farm's packhouse was located near the orchards (within 15 km). Peaches were mainly exported to the UK and EU markets. Precipitation data was gathered from the South African Weather Services (2012).

A total of 428 samples were collected and analysed during four consecutive growing seasons (Table 1; Table 2). During seasons 1 and 4 the farm was visited once during the peak harvesting period, and during seasons 2 and 3 the farm was visited twice, once during the last spray period (one month prior to harvest) and once during the harvest period (Table 2).

Water samples (n = 95; 5 × 1000 ml per site) were collected using a telescopic water sampling arm (1.5 m) (Table 2). Water was collected from holding dams, river, pesticide fill point and at the packhouse. *Prunus persica* L. cv. Oom Sarel samples (n = 60) were collected from the orchard (preharvest) and from the packhouse (postharvest). In the orchard, fruit were collected from five trees from a single orchard block, at four points per tree and three fruit per point. Location of the trees were recorded and visited in subsequent seasons. Five fruit samples of three fruit were randomly collected before and after packing. Transport swabs with Amies medium (Lasec, Johannesburg, SA) were wetted in the transport medium and used to sample a 25 cm^2 area of all contact surfaces according to standard procedures for environmental swab sampling (Public Health England, 2014). Preharvest samples in the orchard included; hands of pickers and crates. Postharvest samples in the packhouse included, hands of workers, processing line (sort- and pack-line), taps (bathroom and wash station) and floors. In the cold room; floors and walls were sampled. All packhouse equipment was recorded as being cleaned daily with water and soap. All samples were transported on ice, stored refrigerated and processed within 24 h (water) to 48 h (fruit) and swabs were processed within one week after collection.

2.2. Hazard characterisation

Water samples (100 ml) were processed for Colilert-18® (Dehteq, SA) analysis as per manufacturer's instructions and incubated at 37 °C. Results were recorded and the most probable number (MPN) of coliforms and *E. coli* were determined.

Further to this water samples (1000 ml), fruit sample rinsates and swab samples were analyzed for the presence of *E. coli* O157:H7, *Listeria* spp., *Salmonella* Typhimurium and *S. aureus* using molecular PCR detection. Water samples (1000 ml) used for molecular detection were filtered through a 0.45 μm nitrocellulose filter. Fruit samples were washed in 500 ml quarter strength Ringer's solution (Merck, SA) amended with 0.02% Tween-80 in an ultrasonic bath for 5 min and subsequently filtered through a 0.45 μm nitrocellulose filter.

Filters and swabs were analysed by placing each into 9 ml tryptone soy broth, shake incubated (100 rpm) at 37°C for 48 h followed by DNA extraction and PCR with negative control, as outlined by Standing, du Plessis, Duvenage, and Korsten (2013) targeting the *UidA* gene of *E. coli* O157:H7 (F: 5′-GCG AAA ACT GTG GAA TTG GG-3′; R: 5′-TGA TGC TCC ATA ACT TCC TG-3′; 252bp amplicon) (Cebula, Payne, & Feng, 1995), the listeriolysin O gene of *Listeria* spp. (F: 5′-AGC TCT TAG CTC CAT GAG TT-3′; R: 5′-ACA TTG TAG CTA AGG CGA CT-3′; 450bp amplicon) (Thomas, King, Burchak, & Gannon, 1991), the long polar fimbriae D gene for the detection of *Salmonella* Typhimurium (F: 5′-TTG CCG GTG GTA CTG ATA GG-3′; R: 5′-TTG CCG GTG GTA CTG ATA GG-3′; 787 bp amplicon) as well as *Staphylococcus aureus* nuclease gene (F: 5′-TTG CAT ATG TAT GGC AAT TGT T-3′; R: 5′-TTT TGC TTG TGC TTC ACT TTT TC-3′; 655 bp amplicon) (Standing et al., 2013). For positive control purposes, PCR reaction mixtures containing DNA extracted from artificially