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An Observational Assessment of Australian Apple Production Practices for Microbial Control

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Key words

Apples, packhouse, food safety controls, microbial risk assessment, risk management

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Highlights

- The variation in wash water sanitary control affected detected *E. coli* levels
- *E. coli and Salmonella* spp. were not detected on apples
- There was low detection rate of *Listeria* spp. on apples
- Interpretation of food safety guidelines for apples varies between packhouses
- Food safety controls applied differently results in variable practices and outcomes

Abstract

Food safety management criteria are often described in general terms rather than specific actions and potentially introduces subjectivity to interpretation and implementation. In the tree fruit sector, management systems would be more useful if developed with specific reference to production and processing practices used. There is insufficient evidence that requirements for the Australian tree fruit industry are appropriate to control foodborne pathogen contamination of ready-to-eat products. Thus, the purpose of this study was to explore industry interpretations of food safety guidelines by describing the application of controls in Australian orchards and packhouses and to evaluate production practices by characterising potential microbial risks in the apple industry, quantifying microbial load in wash water and fruit, and assessing fruit quality as indicators. Thirteen orchards and packhouses across Australia were visited from July 2016 to April 2018 to observe apple orchard practices, packhouse systems, wash water controls, general hygiene and to evaluate the presence of *Escherichia coli*, *Salmonella* spp. and *Listeria* spp. on multiple apple cultivars. The assessment revealed that the inconsistent application of water sanitation resulted in variable control of wash water quality and hygiene, but the prevalence of pathogens on apples was less than 2%. Variation in practices could increase the risk of foodborne illness to consumers if contamination occurs. The Australian apple industry could benefit from a better understanding of effective risk mitigation strategies, consistent industry application of food safety controls and improved evidence of controls achieving desired food safety outcomes.

1. Introduction

Even though growers use good agricultural practices and comply with various food safety standards, transmission of foodborne illness via fresh fruits and vegetables has been identified as an emerging issue in Australia (FSANZ 2020) due to an increased number of foodborne incidents (Butler, Pintar et al. 2016).

Apples are typically consumed raw (APAL 2016) without a processing step to inactivate pathogens; hence, pre- and post-harvest activities need effective management to minimise contamination by microbial pathogens. In the United States in 2014 an outbreak of *listeriosis* from caramel apples, resulting in 35 illnesses with 20% mortality (Angelo, Conrad et al. 2017), alerted the apple industry to potential risks in the supply chain and raised questions concerning control of hazards in orchards and packhouses.

As the requirements for certification of food safety management increase, risk-based evidence of their effectiveness is needed. Fresh fruit microbial risk assessments (MRA) (Duffy and Schaffner 2002, Bassett and McClure 2008, Duvenage and Korsten 2017) and additional studies identified the most likely sources of contamination of fruit as birds (Duffy and Schaffner 2002), animals and water (Suslow, Oria et al. 2003, Park, Szonyi et al. 2012), food handlers (pickers) (Food and Drug Administration (FDA 1998), Food and Agriculture Organisation and World Health Organisation (FAO and WHO 2008), European Food Safety Agency (EFSA 2017)), equipment (FDA 1998, EFSA 2017) and dust (Burnett, Chen et al. 2000, Kumar, Williams et al. 2017). Inadequately sanitised wash water and poor hygiene in packhouses have also been associated with outbreaks (Gibbs, Pingault et al. 2009, Garner and Kathariou 2016). *Listeria monocytogenes* survives on apples (Salazar, Carstens et al. 2016) and, together with *Escherichia coli* O157:H7 and *Salmonella* spp., can grow on damaged apple tissue (Riordan, Sapers et al. 2000, Leverentz, Conway et al. 2003, Allegre, Abadias et al. 2010).

In the Australian apple industry, approaches to MRA to develop risk management, subsequent implementation of controls, and their effect on food safety have not been investigated. Although the prevalence of foodborne pathogens on Australian apples is low (Department of Agriculture, 2020), this study aimed to identify potential gaps in microbial risk management in apples in the context of current orchard and packhouse food safety management systems in Australia. The approach used

was to review current operations and their controls in practice in the Australian apple industry and their potential effect on food safety assessed by hygiene performance and MRA.

2. Materials and methods

2.1. Selection and characterization of study sites

Five orchards (O1-O5) and eight packhouses (P1-P8) in Australia were visited from July 2016 to April 2018 (Table 1). Sites were selected in collaboration with industry experts from Apple and Pear Australia Ltd. (APAL), to represent industry diversity in growing region, size, operational system and practices. All orchards and packhouses were certified to at least one Global Food Safety Initiative (GFSI) benchmarked quality assurance standard e.g. Global G.A.P., Freshcare (Freshcare 2019, GlobalGap 2019) and at least one Australian retailer standard e.g. Woolworths Limited 2013. Various packhouse processing systems and supply chains were assessed to compare operations, food safety management controls and practices.

2.2 Field study design

Orchards 1 to 5 participated in the observational study. However, only growers at orchards 4 and 5 were interviewed. Packhouses 1 to 8 participated in the observational study with packhouses 1 to 6 included in the microbial and quality assessment study.

2.3 Microbial and quality assessment

2.3.1 Sampling scheme

A total of six 500 mL wash water samples and 54 randomly selected apple samples were collected aseptically during production using standard sampling protocols (Taylor, Sofos et al. 2015, AWWA and APHA 2017) and transported to National Association of Testing Authorities (NATA) accredited laboratories by air or road within 24 hours. All samples were maintained and stored at <10°C. At each packhouse, one sub-surface water grab sample from a wash tank was collected and six apple

125 samples (five apples per sample) were collected at three points – pre-wash, post-wash and post-
126 controlled atmosphere (CA) storage. Three samples per sampling point (15 apples in total) were sent
127 for microbial testing. Three samples (of 5 apples) were assessed for quality: soluble solids
128 concentration (Brix°) and firmness (kgf) indicating ripeness, dust caking, calyx cracking and physical
129 damage (unhealed wounds, bruising, hail, sunburn, russet) (HIA and APAL 2016) to determine any
130 relationship with microbial contamination. Apple grade and provenance were recorded ((not
131 reported). Wash water treatment is reported (Table 2). Points of collection and apple variety
132 sampled were dependent on the logistics and availability at each packhouse. Consequently, the
133 study included seven apple cultivars.
134
135 *2.3.2 Microbial analysis methods*

136 For each sample, apples (n = 5) were cut longitudinally into eight pieces then horizontally in 1 cm
137 sections and manually mixed for 1 min in a stomacher bag. Portions were removed for preparation
138 of first dilutions by Stomacher (Model 400 Circulator, Seward, Norfolk, England) for 2 min: 10 g for *E.*
139 *coli* in 90 mL Buffered Peptone Water (BPW) (Oxoid Australia), 25 g for *Salmonella* spp. in 225 mL
140 BPW and 25 g for *Listeria* spp. in 225 mL Half Fraser Broth (Oxoid Australia).
141
142 Analysis of apple samples for *E. coli* followed ISO 16649-2 pour plate method modified by using
143 ChromID medium (bioMerieux Perth, Melbourne) incubated at 37°C for simultaneous enumeration
144 of *E. coli* and coliforms with a detection limit of 10 CFU/g. The presence/absence of *Salmonella* spp.
145 and *Listeria* spp. in samples was assessed by SureTect™ PCR (ThermoScientific™ Adelaide) providing
146 a detection limit of 0.04 CFU/g. Water samples were tested for indicator bacteria by Australian
147 Standard/New Zealand Standard (AS/NZS) 4276.5 2007 for coliforms and AS/NZS 4276.7 2007 for
148 thermotolerant coliforms and *E. coli* using membrane filtration with a detection limit of 1 CFU/100
149 mL.

150 *2.3.3 Quality assessment*

151 Assessment of apple quality was done on-site immediately after sample collection. Apples in each
152 sample were individually assessed against industry guidelines (HIA and APAL 2016). 'Dust caking' was
153 defined as the presence of dirt around the stem or calyx. 'Cracking' was used to describe visible
154 cracks around the stem end, calyx or on the skin (USDA 2002). Evidence of physical damage –
155 bruising, hail damage and unhealed wounds caused by pests or stem puncture – was noted. Brix
156 values were measured using a refractometer and firmness was measured using a fruit penetrometer
157 with 11 mm probe by quality control staff at each packhouse. Whilst equipment manufacturers
158 varied between packhouses, the same principles for measurement were used.

159

160 ## 2.4 Observational study

161 Sites were observed from July 2016 to April 2018 for factors contributing to foodborne pathogen risk
162 (Brackett 1999, FAO/WHO 2003, Suslow, Oria et al. 2003). Food safety management in orchards and
163 packhouses was characterised and assessed based on the systematic approach of Luning and Bango
164 et al. (2008) and modified to include the most likely microbial risk factors to apple production. Three
165 factors of the business environment were included for context (Kirezieva and Nanyunja et al. 2013),
166 ten items of food safety control (Luning and Bango et al. 2008) and four items of assurance activities
167 (Luning and Marcelis et al. 2009) as indicators of microbial performance (Jacxsens and Kussaga et al.
168 2009) were included to obtain a snapshot of industry practices and their efficacy. A checklist
169 developed for this purpose is provided as supplementary material. The study had two components:
170 semi-structured interviews and a hygiene gap audit, i.e. observations of the degree of conformance
171 to best practice based on FDA 1998, FAO/WHO 2003 and the Harmonised Australian Retailer
172 Produce Scheme (HA Ltd. 2016)

173

174 *2.4.1 Semi-structured interviews*

175 Growers (2), operations (7) and quality assurance (8) managers were interviewed about how
176 microbial hazards and risk mitigation strategies were identified and prioritised (food safety policy),
177 what food safety controls were considered the most important, verification of microbial control
178 (food safety assurance), and about challenges in implementing food safety controls. Interviewees
179 were given time during interviews to explore these topics and responses were recorded. If
180 information was missing or unclear on subsequent review, interviewees were later contacted by
181 phone or email for clarification. Responses were scored 1 – 'poor' or 'did not meet', 2 – 'average' or
182 'partially met' and 3 – 'good' or 'met' based on apparent compliance with guidelines/standards
183 (FAO/WHO 1999, FAO/WHO 2003, FPSC 2019, Freshcare 2019).
184
185 *2.4.2 Hygiene assessment*
186 Preventive measures and intervention processes were assessed by gap audit. These included
187 controls for dust contamination, animal and pest intrusion, personal hygiene (protective clothing,
188 availability, cleanliness and utility of handwashing facilities and restrooms), equipment cleanliness
189 and maintenance, building cleanliness, process flow and waste management. Scores were assigned
190 as described in 2.3.1. When control was inconsistent, for example, one food handler did not wear a
191 hairnet, a score of 2 was assigned.
192
193 ## 2.5 Statistical analysis of data
194 Due to zero variability in data from some packhouses, all data were analysed using a Monte Carlo
195 Kruskal-Wallis test (Kruskal and Wallis 1952). Personnel and building/equipment scores were
196 combined and analysed using the equation score = scored/$score_{max}$ and by analysis of variance
197 (ANOVA) using an asymptotic Kruskal-Wallis nonparametric test with the null hypothesis that all
198 results were equal at 95% confidence. For Brix and firmness, the average measurement of the three
199 samples was calculated before data analysis.
200

The study was approved by Social Sciences Human Research Ethics Committee, Research Integrity and Ethics Unit, University of Tasmania (number H0017183).

3. Results and Discussion

3.1 Microbial and quality assessment

3.1.1 *Wash water*

Two of the six packhouses had < 1 CFU/100 mL *E. coli*, coliforms and thermotolerant coliforms in their wash water (Table 2). The two packhouses that monitored wash water continuously had < 1 CFU/100 mL for *E. coli* and thermotolerant coliforms, but coliforms were detected (Table 2). Packhouses without monitoring protocols in place had high levels of all microbial contaminants (>100 CFU/100 mL) . Packhouses with wash water in compliance with current guidelines for *E. coli* (FAO/WHO 2003, FPSC 2019) used 'town' water treated with an approved sanitiser and daily monitoring (P5 and P6). However, automatic dosing or monitoring systems did not ensure the absence of all indicator bacteria suggesting that at times of high organic load (e.g. leaf litter, dust, apple sunscreen) or microbial load on apples the process could allow survival of *E. coli*. When surface water without sanitiser (P3) or rainwater without monitoring (P4) were used high levels of the target bacteria were observed, indicating a potential risk of apples contaminated with pathogens (FDA 1998, Brackett 1999).

3.1.2 *Apples*

Sample collection points in the processing stage were more varied than anticipated (e.g. some pre-wash samples were fresh from the orchard or out of storage), however, apparently this did not affect microbial loads on apples as *E. coli* and *Salmonella* spp. were not detected in any samples

collected (Table 2). Detection rates of *E. coli* in similar surveys are generally low (Abadias, Usall et al. 2008, van Dyk, de Bruin et al. 2016), Duvenage and Korsten (2017), (De, Li et al. 2018), although Abadias *et al.* (2006) found 8.3% of apples sampled from the orchard and 13.9% post-packing had low levels of contamination. Surprisingly, we did not find *E. coli on* apples sampled from the contaminated wash water. This may indicate a low transfer rate as various studies (Pahl, Telias et al. 2013, Won, Schlegel et al. 2013, Xu, Pahl et al. 2015) report a limited relationship between bacterial counts in irrigation water and contamination on produce.

No S*almonella* spp. were detected in apples sampled in this study (Table 2). Detection of S*almonella* spp. on tree fruit varies (Abadias, Canamas et al. 2006, Gomba, Chidamba et al. 2016). Abadias et al. (2006) found no S*almonella* spp. in a whole supply chain survey of 216 apple samples in Spain collected after CA storage but Gomba et al. (2016) found nearly 5% contamination in tree fruit samples taken from 225 orchard and packhouse locations in South Africa . In the latter study, when either irrigation or wash water was positive for *Salmonella* spp., the pathogen was also detected on fruit. Although not a tree fruit, a survey of 117 field and packhouse tomato samples (van Dyk, de Bruin et al. 2016) failed to detect any S*almonella* spp. This suggests that if the water is clean, the fruit will also be clean.

In this study, *Listeria* spp were detected (at <10 CFU/g) in only one apple sample before washing (Table 2). *Listeria* spp. were not detected in the same batch of apples sampled post-wash and no indicator bacteria were detected in the wash water. The wash water was treated with 30 ppm chlorine and well monitored. There are few non-outbreak related surveys for *Listeria* spp. for tree fruit (Abadias, Usall et al. 2008, Uchima, de Castro et al. 2008, Duvenage and Korsten 2017). A low detection rate of 1.8% (1/54) in this study was similar to that found for peaches (Duvenage and Korsten 2017) and consistent with other fresh produce surveys (FSANZ 2010). Given the source of the samples, the contamination might have been due to a dust-affected bin.

Although differences for quality parameters were found between packhouses, no significant consistent differences were apparent between fruit quality parameters and microbial detection. However, dust caking and damaged fruit were observed in all packhouses (Table 3) adding to the potential for contamination (Riordan, Sapers et al. 2000, Kenney, Burnett et al. 2001, Kumar, Williams et al. 2017) and accentuating the need to determine risk points in the operations and mitigations.

3.2 Observational study

Food safety management practices were explored to evaluate producer understanding and interpretation of quality control requirements and assess the potential for microbial risk. Results of interviews and hygiene assessment are combined in Table 4. 'Food safety controls' (FSC) refer to preventive activities that were documented policy at the interviewees' businesses and observed in practices. 'Food safety assurance' refers to verification of sanitiser use and microbial testing.

3.2.1 *Contextual risk factors*

Water source

Six packhouses used town water supply for washing operations, one site used rainwater and one site accessed river water for apple washing, resulting in scores 'good', 'average' and 'poor' respectively (Table 4). Irrigation water included farm dams, river and greywater, all of which are known sources of pathogens that can cause direct or indirect contamination (FDA 1998, Park, Szonyi et al. 2012). The two orchards scored were assigned 'average' because there was little observational evidence of interventions to lower risk of water-related contamination (Table 4). However, the farm dam at one orchard, filled by rainwater, was lined with plastic to reduce contamination from sediments after rainfall. While under-canopy spray irrigation and fertigation were used, which minimised direct fruit contact and thus risk from pathogens in water (FDA 1998), overhead sprinklers were used for foliar

278 spray application and cooling apples. Dropped fruit may allow contact with irrigation water and soil,
279 with a risk of associated pathogens transferring to fruit surfaces (Duffy and Schaffner 2002). Orchard
280 and packhouse policies excluded dropped fruit at harvest, but packhouse managers indicated that
281 control was difficult. Another unexpected pathway for contamination from water was the use of
282 irrigation water to dampen sack covers on harvested apples for the prevention of sunburn.
283
284 3.2.2 *Food safety controls*
285 *Dust and soil*
286 Dust or soil control was poor in the orchard because dirt roads (Table 4) increased the potential for
287 fruit contamination as demonstrated for *Salmonella* spp. and tomatoes (Kumar, Williams et al.
288 2017). Growers scored a 'good' rating when they sought to reduce cross-contamination to apples by
289 using single-use plastic bin liners or daily cleaning/sanitising of picker bags. However, one grower
290 said, "crates are often returned from wholesalers uncleaned, so fruit gets contaminated anyway".
291 Three packhouse managers raised concern over dust and soil contaminating fruit and harvest bins,
292 with one stating "I work on the principle of remove the dirt, remove the problem". Splash-back from
293 muddy bins could result in cross-contamination to apples (Allende, Castro-Ibanez et al. 2017). Dirty
294 bins and apples also increase the organic load in dump and wash tanks, reducing the effectiveness of
295 sanitisers (Suslow 1997, FDA 1998) and increasing potential for pathogen survival and cross-
296 contamination between fruit (Gil, Selma et al. 2009).
297
298 *Animals and pests*
299 Although intrusion of domestic animals was controlled, kangaroo droppings and birds were seen in
300 orchards so an 'average' rating was assigned (Table 4), because the potential for pathogen transfer
301 from faeces to bins during harvest was demonstrated. Native animals, rodents and sheep seen
302 grazing adjacent to orchards were potential sources of direct and indirect faecal contamination to
303 farm dams (Suslow, Oria et al. 2003, Park, Szonyi et al. 2012). The increased use of netting in

orchards to modify climatic conditions may reduce the likelihood of bird damage thus lowering microbial risk. When assessing waste management, less decaying fruit was observed in orchards where under-tree debris was swept away to mitigate risk of rodent activity. As rodents can carry and shed pathogens (e.g. *Salmonella* spp.) (Meerburg, Singleton et al. 2009, Kilonzo, Li et al. 2013), reducing their prevalence through good orchard hygiene would be expected to decrease the risk of contamination of fruit on trees.

Building design and process flow

In all orchards, fruit was temporarily held in open, general storage sheds (assigned 'average' rating), providing points for pest harbourage and cross contamination to harvested apples. One site used a lean-to exposed to prevailing winds (assigned a 'poor' rating) (Table 4).

Harvested apples delivered to packhouses are placed in water dumps to float the apples through wash water tanks. Conveyor travel through drying tunnels, waxing and graders follows in various order, after which they are clean and move to the packing lines. Apple packing lines are dry areas where the risk of pathogens like *L. monocytogenes* is lower (Sutherland, Miles et al. 2003). The operational workflow in two packhouses with high ratings facilitated movement from packing (low contamination) to bin receival (high contamination) to prevent cross-contamination of finished product and compliance was enforced (Table 4). Low and high contamination areas were not demarcated at four packhouses where the workflow was in the opposite direction. In two packhouses direction of movement was *ad hoc*, lowering the rating assigned for this feature (Table 4). Failure to control people movement through facilities increases the risk of cross-contamination of finished product from dirty and wet areas (Sutherland, Miles et al. 2003, UFPA 2013).

Large packhouses offering centralised processing facilities with longer distances between fruit receival and despatch are reported to lower the likelihood of microbial contamination in wash and

packing areas from dust, birds and pests (Suslow, Oria et al. 2003). However, if these facilities did not use internal walls as physical barriers to prevent cross-contamination by air, water or traffic flow they were scored "average" (Table 4).

Washing areas in all packhouses operated at ambient temperature where apples were held for up to 12 hours. Growth of pathogens, if present, would be unlikely due to low pH of apples (Wu, Gao et al. 2007) but bruised or wounded fruit that is not removed from the line can provide environments conducive to growth (Dingman 2000, Glass, Golden et al. 2015). Storage conditions in cool rooms, typically 0-4°C and from 0-2°C under CA, inhibit growth of pathogens such as *Salmonella* spp. (Jay, Davos et al. 2003) and *E. coli*, although *L. monocytogenes* can survive for 5 months during cold storage (Macarisin, Sheth et al. 2019).

Building and equipment hygiene

Observation of dirt accumulation on bins, crates, brushes, grading cups and conveyor belts indicated the potential for equipment to contaminate apples (Table 4). Soil can harbour microbes (Ailes, Leon et al. 2008, FPSC 2019) and can be a source of cross-contamination in the packhouse (FDA 1998, Harris, Farber et al. 2003, Suslow, Oria et al. 2003, FPSC 2019), increasing risk of fruit contamination (Gagliardi, Millner et al. 2003). UFPA (2013) and FPSC (2019) recommend that equipment and facilities should be designed, cleaned and sanitised to prevent cross-contamination and development of niches where pathogens can survive.

Only two packhouses cleaned their wash lines daily but procedures varied to include wipe down, air hosing or steam cleaning (Table 4). The effect on risk reduction of these practices is unknown because cleaning verification was infrequent. Ineffective sanitation contributes to fruit contamination and outbreaks (Gagliardi, Millner et al. 2003, Gibbs, Pingault et al. 2009, Garner and Kathariou 2016) thus, verification of cleaning and sanitising is important for risk mitigation. Pre-

356 sizers (water flume graders), although vacuum cleaned weekly were only emptied and scrubbed
357 every 12 to 18 months, so biofilms could form and be a source of pathogens as occurs in other food
358 sectors (Kumar and Anand 1998, Ryu and Beuchat 2005).
359
360 Hygiene scores at each packhouse ranged from 'poor' to 'good' (Table 4) indicating that some
361 controls were well managed, and others were neglected, highlighting the challenges in consistently
362 maintaining good hygiene. There was evidence that cleaning schedules, procedures and their
363 compliance need review in some establishments. Similarly, assessment of waste management
364 showed that wastewater, general rubbish, and organic waste disposal was compromised. Water
365 pooling on floors was observed at two packhouses potentially resulting in splash-back to product,
366 transfer of contaminated water through the facility and increased risk of contamination to fruit and
367 equipment (FDA 1998, DoA W.A. 2002, Suslow, Oria et al. 2003) from pathogens such as *L.*
368 *monocytogenes* (Sutherland, Miles et al. 2003).
369
370 One packhouse used ozone-generating systems to clean surfaces and air in CA rooms. Use of ozone
371 in storage and packing facilities is well established for postharvest disease control (Smilanick 2003).
372 However, verification of its effectiveness for control of foodborne pathogens would be of interest to
373 the fresh fruit industry.
374
375 *Food handlers*
376 Food safety training and staff supervision was found to present a challenge to the industry. Hand
377 washing and use of antiseptic are required by Australian Food Safety Standards (FSANZ. 2000) but,
378 as indicated by the ratings given (Table 4), consistent compliance was problematic despite
379 appropriate hygiene policies being advocated. One grower commented "you can't be certain they do
380 it" and "pickers don't think of themselves as food handlers". This suggests that not all pickers
381 appreciated that direct hand-fruit contact introduces risk of contamination and that good personal

hygiene can reduce contamination (Brackett 1999). Other barriers for food handler hygiene were handwashing facilities or hand sanitiser not being available in all portable toilets in orchards. Gloves were not always worn, particularly in the packhouse, and when they were worn in orchards, they were sanitised only daily. Food handlers can affect the likelihood of contamination of fruit (Brackett 1999), thus, industry-consistent personal hygiene could increase certainty of control.

Only half the packhouses paid attention to use of protective clothing, suggesting differences in risk perception. Although Australian guidelines (FPSC 2019, Freshcare 2019) allow grower/packer discretion for glove use, as does the USA (FDA and DoHHS 2016), failure to enforce protective clothing protocols is inconsistent with best practice (FPSC 2019). The perception of the food safety 'climate' in the facility could explain less stringent control at some sites (De Boeck, Jacxsens et al. 2015). Based on poor ratings at some sites and because food handlers have been implicated in outbreaks (Machado-Moreira, Richards et al. 2019) the lack of validation, verification and monitoring of personal hygiene controls and absence of management tools to measure FSC objectives are important issues requiring attention.

Wash water

Sanitiser use in postharvest water is critical to mitigate microbial risk. The primary purpose is to prevent contamination of water and cross-contamination of apples should pathogens be introduced from apples and bins (FDA 1998, Bassett and McClure 2008). This study provided evidence of variation in wash water sanitary control and its verification (Table 2). Seven packhouses sanitised dump and wash water, consistent with best practice. Four packhouses used chlorinated pre-sizers – three with manual monitoring, one with automatic monitoring. Four packhouses washed apples before and after storage, including the site without water treatment. Multiple washes increase risk management requirements, and thus, the potential for failure. The failure of some packhouses to

control their wash water quality suggests that further education and training of packhouse managers in risk assessment is needed.

Dump and wash water temperatures were not controlled or monitored in any packhouses. Apples were sometimes washed straight from the orchard when fruit temperature was high. Apple surface temperatures of 27°C were reported at one packhouse and one grower said fruit can reach up to 45°C. There is risk of internalisation of pathogens if warm fruit is immersed in colder contaminated wash water (IFT and FDA 2001). Studies indicate that if the core temperature of apples is greater by 13 (Buchanan, Edelson et al. 1999) or 23°C (Burnett, Chen et al. 2000) than wash water pathogen uptake through the calyx is enhanced. Australian guidelines identify this risk in postharvest water use but do not provide specific temperature gradient recommendations (FPSC 2019). Temperature gradients between fruit and wash water and poor sanitary control contributed to an outbreak of *Salmonella* associated with rockmelons in Australia (Munnoch, Ward et al. 2009).

3.2.3 *Food safety assurance*

Verification activity was mostly poor for orchards and in the packhouse environment scores varied (Table 4) due to differences in testing frequency or absence of microbial testing. Certification systems require that verification testing of wash water be conducted at a rate commensurate with the business risk assessment (Freshcare 2019). However, irrigation water quality was generally only assessed annually for certification compliance, indicating inappropriate risk assessment because of insufficient data to understand and describe variation in quality or likelihood of high-risk contamination events (FDA and DoHHS 2016). Cool room cleaning was verified annually by environmental swabbing for *E. coli* and *Salmonella.*

Two packhouse operators assumed that wash water microbial risk was controlled with *ad hoc* addition of chlorine or removal of dirt from bins, but our results showed *E. coli* was present at levels

indicating the potential for pathogen presence in these systems (Table 2). Testing frequency varied at packhouses with no obvious link to the control system used. For example, only two packhouses tested dump water annually for *E. coli.* Curiously, testing town supply source water was common but untreated sources of water were not tested. Testing wash water to increase knowledge of system performance, rather than town supply, would be a better use of resources because data analysis results could be used in risk mitigation decisions.

Microbial testing of fruit was variable with two packhouses doing no testing (assigned a "poor" rating), three packhouses testing one to two times per year ("average" rating) and three packhouses testing four times/year (good rating) (Table 4). Apples were analysed for *E. coli, Salmonella* spp.and *L. monocytogenes.* Australian retail chains have applied quality standards to apple packhouses that include microbiological criteria and fruit verification requirements (Woolworths Limited 2013, Coles Supermarkets Australia Pty. Ltd. 2016). This has encouraged implementation of MRA and preventive actions (Premier and Ledger 2006). However, there was little evidence of real-time responses to changes in risk level. The only example identified in this study was increasing the number of food handlers removing damaged fruit after hail, a potential avenue for contamination (Dingman 2000, Glass, Golden et al. 2015).

Half the packhouses verified equipment cleaning by implementing an environmental monitoring programme; but frequency of testing ranged from one to four times per year. A 'good' rating was given for more frequent verification as it provides greater assurance that microbial hazards are controlled. Samples were analysed for coliforms, *E. coli,* coagulase positive staphylococci, *Salmonella* spp. and *L. monocytogenes* (1 packhouse) or *E. coli* and *Listeria* spp. (3 packhouses).

Assurance activities did not correlate with the level of potential risk observed, in that less developed management and technology generally require more verification to manage risk (Luning, Marcelis et

al. 2009, Kirezieva, Nanyunja et al. 2013). In this study, packhouses with less sophisticated operational systems and/or less sanitary controls did the least verification, thereby preventing use of data to assess risk and make appropriate changes. Low verification activity might be due to lack of understanding of the benefits conferred, cost, or logistics associated with sample analysis.

4. Conclusion

The packhouses participating in this study had low prevalence of food-borne pathogens on apples. However, observational assessment provided evidence of very inconsistent application of hygiene controls. Although correlation between the level of hygiene control and the presence of pathogens could not be discerned and there was no relationship between microbial water quality and pathogen prevalence on apples, despite variable levels of sanitisation, an indication was obtained that pathogens enter the packhouse from the orchard, highlighting the importance of consistent and reliable hygiene control and the need for routine monitoring programs to identify contamination hotspots. Thus, this study raises questions over grower interpretation of standard audit results which are a snapshot in time, in assessing risk and suggests that auditing focused on the details of critical food safety controls like water sanitisation would be valuable.

Inadequately sanitised wash water and equipment can lead to contaminated produce and outbreaks. A low level of system verification exposes customers to risk of foodborne illness if pathogen contamination occurs and can lead to reputational risk to the industry. Where interviewee responses did not differ from observations, there was good understanding about food safety control, and sound technical understanding of wash water sanitisation resulted in good microbial control of wash water. In general, however, improved knowledge and application of risk assessment methods would benefit the industry.

Despite orchards and packhouses using analogous certification standards, food safety controls and verification (assurance) activities varied, indicating insufficient knowledge of system performance. This presents an opportunity for the industry to further investigate the effect of different approaches to food safety management and thus acquire the knowledge needed to ensure consistent outcomes are achieved from implemented controls. In this study, overall food safety evaluated by hygiene audits and microbial assessment of wash water and apples, varied from 'poor' to 'good'. This highlights the importance of more specific guidelines based on risk assessment for apples that can be easily and consistently interpreted. The lack of evidence- and outcome-based requirements in standards may be a barrier to improving industry consistency because they allow individual interpretation. Although this was a small, non-systematic study it provided valuable insight for the Australian apple industry on the range of current practices, gaps in microbial hazard control and evidence of control. While further studies should focus on the effect that variations in practices have on microbial risk, this study has highlighted the need for tools to measure food safety management performance and assist risk-based decision-making.

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719

Table 1. Characterisation of the orchard (O) and packhouse (P) study sites

Study site	Growing region	Organisational structure	Size[1]		
			S	M	L
O1	Region A, South Australia	Independent grower	X		
O2	Region A, South Australia	Independent grower			X
O3	Region A, South Australia	Independent grower		X	
O4	Region B, Western Australia	Private family company			X
O5	Region C, Western Australia	Private family company			X
P1	Region D, Victoria	Owner/operator		X	
P2	Region D, Victoria	Joint owner/operator			X
P3	Region E, Victoria	Owner, operator	X		
P4	Region A, South Australia	Owner, operator		X	
P5	Region A, South Australia	Cooperative			X
P6	Region C, Western Australia	Owner/operator		X	
P7	Region B, Western Australia	Owner/operator		X	
P8	Region B, Western Australia	Private family company		X	

720 [1] Orchard size (ha): S small <100, M medium 100-200, L large >200

721 Packhouse capacity (number of bins): S small <10,000, M medium 10,000-25,000, L large >25,000

722 **Table 2. Wash water treatment type, target level, monitoring frequency and results of a snapshot of bacterial indicator organisms and pathogens in wash**
723 **water (n = 6) and apple (n = 54) samples; each sample is five apples, from six Australian packhouses.**

Site	Treatment	Target	Monitoring	Wash water microbial load[a]			Apple microbial load								
				Coliforms	Thermotolerant coliforms	*E. coli*	*E. coli*[b]			*Listeria* spp.[c]			*Salmonella* spp.[c]		
							Pre-wash	Post wash	Post CA	Pre-wash	Post wash	Post CA	Pre-wash	Post wash	Post CA
P1	Peroxitane	ORP 420-580	Continuous	200	<1	<1	<10	<10	<10	ND	ND	ND	ND	ND	ND
P2	Chlorine dioxide	0.1-0.3 ppm	Continuous	≈12	<1	<1	<10	<10	<10	ND	ND	ND	ND	ND	ND
P3	None	none	None	5,300	4,000	4,000	<10	<10	<10	ND	ND	ND	ND	ND	ND
P4	Nylate^	ORP 650	None	11,000	200	200	<10	<10	<10	ND	ND	ND	ND	ND	ND
P5	Chlorine	5 ppm	Daily	<1	<1	<1	<10	<10	<10	ND	ND	ND	ND	ND	ND
P6	Chlorine	30 ppm	Daily	<1	<1	<1	<10	<10	<10	D x 1	ND	ND	ND	ND	ND

724 P packhouse, ORP oxidation reduction potential,l ppm parts per million, ^bromo chloro dimethyl hydrantoin
725 [a]colony forming units per 100 mL, [b]colony forming units per g, [c]detected (D)/not detected (ND) in 25 g. Apple results n = 3 at each sampling point.

726

Table 3. Results of apple quality measured by sugar content and firmness, and assessed by features potentially related to microbial contamination. Brix and firmness results are averages (n=15) of triplicate samples of five apples. Dust caking and physical damage results are the number of positive apples from n=15.

Site	Brix (%)			Firmness (kgf[b])			Dust caking			Physical damage (major)		
	Pre-wash	Post-wash	Post-CA[a]	Pre-wash	Post-wash	Post-CA	Pre-wash	Post-wash	Post-CA	Pre-wash	Post-wash	Post-CA
P1	13.3	13.6	13.9	8.4	8.7	8.7	0	1	0	1	2	0
P2	12.2	14.3	12.2	8.3	9.3	8.3	0	0	1	0	1	1
P3	12.8	12.0	12.0	7.9	7.4	7.4	0	1	0	2	2	1
P4	13.6	13.6	13.6	5.9	5.9	5.9	0	0	1	1	1	1
P5	16.2	15.7	15.1	9.3	10.0	9.4	2	0	0	0	2	3
P6	13.0	13.9	12.3	8.6	8.6	8.0	1	0	0	0	0	3

727 [a]Controlled atmosphere [b]kilogram force

728 **Table 4. Assessment of food safety management according to observational analysis and semi-structured interviews. Scores of 'poor', 'average' and 'good'**
729 **were based on compliance assessed against guidelines. Numbers represent the number of orchards or packhouses in each assessment category. Two**
730 **orchards and eight packhouses participated in the semi-structured interviews.**

Feature	Orchards			Packhouses		
	Poor	Average	Good	Poor	Average	Good
Context						
Food safety policy	-	2	-	-	4	4
Relationship with suppliers	-	2	-	-	8	-
Water source	-	2	-	1	1	6
Food safety controls						
Dust/soil	1	1		2	3	3
Animal/bird/pest intrusion	-	1	1	2	3	3
Building design	1	1		1	4	3
Process flow	na	na	na	1	4	3
Building hygiene	1	-	1	3	2	3
Equipment hygiene	-	1	1	2	3	3
Waste	-	-	2	-	6	2
Cleaning and sanitation	-	1	1	-	6	2
Food handler hygiene	1	1	-	1	3	4
Toilet/handwashing facilities compliance	-	-	2	-	2	6
Food safety assurance verification						
Irrigation/wash water:						
Chemical	na	na	na	2	4	2
Microbial	2	-	-	4	2	2
Fruit microbial testing	1	1	-	2	3	3
Environmental swabbing program	-	2	-	4	3	1

731 na not applicable

Highlights

- The variation in wash water sanitary control affected detected *E. coli* levels
- *E. coli and Salmonella* spp. were not detected on apples
- There was low detection rate of *Listeria* spp. on apples
- Interpretation of food safety guidelines for apples varies between packhouses
- Food safety controls applied differently results in variable practices and outcomes

Conflict of Interest and Authorship Conformation Form

- All authors have participated in (a) conception and design, or analysis and interpretation of the data; (b) drafting the article or revising it critically for important intellectual content; and (c) approval of the final version.

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