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Pre-and post-harvest practices of smallholder farmers in rural KwaZulu-Natal, South Africa: Microbiological quality and potential market access implications

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ABSTRACT

Increasing demands for "minimally processed" fresh produce create a potential market niche in high-value markets for South African smallholder farmers. Fresh produce not meeting required microbiological quality criteria causes consumer health concerns and therefore limits market access. This study determined selected smallholder farmer pre-and-post-harvest practices and their potential contributions to the microbiological quality of "minimally processed" fresh produce and thus potential implications for market access. Survey results indicated that most smallholder farmers used animal manure as fertilizer (74%) and that their knowledge of potential contamination sources was poor. Microbiological analysis showed that a number of irrigation water samples did not meet WHO recommendations for fecal coliform levels and exceeded the South African standard for *E. coli* for irrigation water applied to "minimally processed" fresh produce. Lettuce, parsley, carrots, and spinach collected over at least 3-months were frequently of unsatisfactory quality with respect to total coliform (ranging from 130 to 79000 MPN/g) and *E. coli* levels (ranging from 2.2 to 49 MPN/g) according to South African Department of Health recommendations. While surfaces within the processing chain harbored *E. coli*, *Salmonella* spp. was not detected on surfaces, in fresh produce or irrigation water samples analyzed. Antibiotic susceptibility patterns of 155 randomly selected *E. coli* isolates from fresh produce and irrigation water were determined by using a disk diffusion method, showing high percentages of resistance for streptomycin (95%) and amoxicillin-clavulanic acid (32%). In addition, the ability of *E. coli* isolates from fresh produce to form biofilms was shown. The identification of antibiotic-resistant and biofilm forming *E. coli* from fresh produce and within the production environment highlights the importance of pre-and post-harvest hygiene practices, especially if smallholder farmers intend on supplying high-value markets.

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1. Introduction

Good quality fresh produce is an essential component of a healthy human diet (Hussain & Gooneratne, 2017). For South Africa, fresh produce is important due to its potential health benefits, availability, and its apparent ease of production. A large proportion of the South African population comprises of so-called YOPI (Young, Old, Pregnant, and Immune-compromised) individuals that are more susceptible to infections. This is partly due to the high prevalence of TBC as well as HIV and the widespread use of anti-retroviral treatment (Day & Gray, 2016; Oni et al., 2015). Hygienically safe fresh produce is therefore an essential dietary requirement. In South Africa, a majority of rural households practice smallholder farming, which is mainly characterized by farming on plots < 2ha (Aliber, Kirsten, Maharaj, Nhlapo-Hlophe, & Nkoane, 2006). It provides fresh produce for families and can contribute to income generation given that produce surplus is available and safe for consumption and of good quality to be marketed (Thamaga-Chitja & Hendriks, 2008).

Fresh produce is a natural habitat for a large array of microorganisms (Gorni, Allemand, Rossi, & Mariani, 2015). However, contamination of fresh produce with foodborne disease causing organisms such as pathogenic *Escherichia coli* strains and *Salmonella* spp. arising along the farm to fork continuum can render fresh produce unsafe for consumers (Berger et al., 2010; Holvoet,

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Sampers, Seyynaeve, Jacxsens, & Uyttendaele, 2015). Potential sources of contamination include pre-harvest practices such as using fresh manure as fertilizer and poor quality irrigation water as well as numerous post-harvest practices, including handling and packaging of produce (Francis et al., 2012; Harris et al., 2003; Olaimat & Holley, 2012). "Minimally processed" fresh produce such as lettuce is often consumed raw, thus posing a risk to consumers if contaminated with pathogenic microorganisms (Holvoet et al., 2015). This is even more concerning, if foodborne disease causing pathogenic bacteria present on contaminated produce display antibiotic resistance, as such pathogens might be more difficult to treat using key antibiotics (EFSA, 2018).

Microorganisms tend to form so called biofilms on both abiotic and biotic surfaces such as processing tables, farming equipment, and plants (Rayner, Veeh, & Flood, 2004; Srey, Jahid, & Ha, 2013). These biofilms represent a physical state of resistance towards cleaning and disinfection displayed by microorganisms (Costerton, Geesey, & Cheng, 1978) and were involved in an estimated 65% of all microbial related diseases (Joo & Otto, 2012). Although fastidious pathogens do not often form biofilms under nutrient limiting environmental conditions, produce contaminated between farm and fork present an opportunity for pathogen attachment and biofilm formation if nutrients become available due to damaged or injured surfaces of fresh produce (Jahid & Ha, 2012). Biofilms present on fresh produce, contact surfaces or equipment within pre- and post-harvest processing are generally difficult to remove and serve as a potential means of cross-contamination, further highlighting why biofilms represent a potential threat to food safety and quality (Jahid & Ha, 2012; Srey et al., 2013).

Conventional fresh produce processing methods are generally assumed to extend the shelf life of food products because they limit the abundance of microorganisms by controlling their growth, while the limited processing of ready-to-eat "minimally processed" fresh produce often renders this more perishable (Rico, Martin-Diana, Barat, & Barry-Ryan, 2007). The pre- and post-harvest practices employed by South African smallholder farmers are mainly traditional methods centered on good farming principles, passed down from generation to generation (Louw, 2013; van der Heijden & Vink, 2013). However, these methods are frequently not carried out in the most efficient manner (DAFF, 2012). Therefore, most South African smallholder farmers require more information on hygienic production and good agricultural practices, such as emphasized by the United States Food and Drug Administration (FDA, 2008) and the Food and Agriculture Organization of the United Nations (FAO, 2003). It is contamination through inappropriate methods, which can lead to smallholder farmers producing fresh produce not meeting market standards (Mdluli, Chitja, & Schmidt, 2013). Consequently, this decreases their potential to access high value markets to generate additional income.

The aim of this study was to assess the pre- and post-harvest hygiene practices of smallholder farmers and to investigate the potential impact on the microbiological quality of "minimally processed" fresh produce and thus market access challenges. This was done by sampling and analyzing irrigation water as a potential source of contamination, fresh produce supplied by smallholder farmers, and surfaces involved in the production and post-harvest processing of fresh produce for the presence of selected hygiene indicator organisms and the potential pathogen *Salmonella* spp. In addition, the antibiotic resistance patterns of *E. coli* isolates from farming samples and their biofilm formation potential were determined. Furthermore, the pre-and post-harvest practices employed and their relative impacts were determined by key informant interviews and semi-formal questionnaires.

2. Methods and materials

2.1. Study site

Data collection tools included key informant interviews with staff of the Marianhill Agri-hub, a local organic farming non-governmental organization (NGO) and questionnaires administered to 80 out of 500 subsistence farmers all affiliated with the NGO, who volunteered to participate. The staff interviews were carried out at the headquarter office of the Agri-hub, located at the Marianhill site. The fresh produce supplied by the Marianhill Agri-hub is labelled "organically produced" but not "certified organic" as organic certification has not been obtained by the organization. The Marianhill Agri-hub consists of four Agri-hub's, namely: Hambanathi, uMbumbulu, Marianhill and Cliffbux (situated in Cliffdale), all located in the province of KwaZulu-Natal, South Africa (Fig. S1). Questionnaires were used to collect information that would provide insight into the attitudes and behaviors of the smallholder farmers selected for the study. This study included both trained (49/80) and untrained farmers, as well as farmers with different educational backgrounds (10/80 without any formal education and 13/80 with tertiary education). In principal, all farmers are supplying the Agri-hub, but only when surplus produce is available. The questionnaires (Supplementary information), prepared in English and translated into isiZulu on site, provided insight into farmers' pre- and post-harvest hygiene practices.

2.2. Data analysis

Data was coded, captured, and analyzed using the IBM SPSS (version 24, 2016) statistical package. Frequency tabulations and mean value calculations for selected pre- and post-harvest practices were carried out to describe samples. The significance of relationships between farmer pre-and post-harvest hygiene practices and selected categorical socio-economic variables were evaluated by the Pearson Chi- Square test. P-values < 0.05 were considered as significant.

2.3. Sample collection for microbiological analysis

Produce (January to April 2016) and irrigation water (February to April 2016) samples were collected and analyzed once a month. Fresh produce samples (lettuce, parsley, spinach, carrots) were randomly collected from the Marianhill Agri-hub, which therefore represent the fresh produce supplied by different smallholder farmers from the four mentioned districts. Irrigation water samples representative of water sources frequently used for irrigation by smallholder farmers were randomly collected and analyzed. These included samples from a Pond (this served as a substitute for borehole water, which was not available at the time of sampling), a rainwater collection device, stream water from a farmer's yard, and a communal river (Mnini District, uMbumbulu). Municipal tap water was collected from the Marianhill Agri-hub facility (Figs. S2A–F).

2.3.1. Water sampling

Irrigation water samples were obtained from the locations specified above between 8am and 12pm using sterile 500 ml Schott bottles. Generally, water samples were collected at a depth half that of the total, in order to avoid collection of exclusively surface water as well as debris. In the case of free flowing water sources, samples were collected from areas of fast flow. For the collection of municipal tap water, autoclaved bottles were used containing 0.1 ml of a $Na_2S_2O_3$ x 5 H_2O solution (18 mg/ml) per 100 ml sample volume to neutralize free chlorine present.

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