Evaluation of vegetable washing chemicals

Robert Premier
Global F.S. Pty Ltd

Project Number: VG09086

VG09086

This report is published by Horticulture Australia Ltd to pass on information concerning horticultural research and development undertaken for the vegetables industry.

The research contained in this report was funded by Horticulture Australia Ltd with the financial support of the vegetables industry.

ISBN 0 7341 3028 7

Published and distributed by:
Horticulture Australia Ltd
Level 7
179 Elizabeth Street
Sydney NSW 2000
Telephone: (02) 8295 2300
Fax: (02) 8295 2399

HAL

Horticulture Australia

Evaluation of vegetable washing chemicals (VG09086)

Final report for HAL project

(June 2012)

Robert Premier

Global F.S. Pty Ltd

Evaluation of vegetable washing chemicals

Final report for HAL project VG09086

By: Robert Premier

Project Leader
Dr Robert Premier
Global F.S. Pty Ltd
Ph: 0418317786
Email: robert.premier@consultant.com

Scope of the Report
This report presents the key findings and a summary of the work conducted in Victoria by the project team to evaluate various wash water treatments available to commercial vegetable growers to reduce the incidence of food borne illness and postharvest diseases.

Funded By: Horticulture Australia Limited using the vegetable levy and matched funds from the Australian Government.

Evaluation of vegetable washing chemicals

Contents

Acknowledgements:

The project team would like to acknowledge the support and input from all those that have contributed to the project including the participating growers who wish to remain anonymous. In addition the team would like to acknowledge the School of Land and Environment, University of Melbourne, Sandy Clarke from the University of Melbourne statistical consulting centre and Dr Marie-Astrid Ottenhof for reviewing the manuscript.

1. Media Summary

Vegetables, including leafy vegetables, are an important component of a healthy diet; they contain nutrients that are now known to be beneficial for the health of consumers. These nutrients are often referred to as phytochemicals or phytonutrients. They have a range of actions on the human body through their interaction at the cellular level. There is sufficient information in the literature to show that they protect consumers from heart disease, inflammation, cancer formation and many other chronic diseases. The anti cancer properties of phytonutrients can be due to a number of interactions, including the induction of phase I or II detoxification enzymes, modulation of phase I and other enzyme activities, antioxidant activity, electrophile scavenging activity, inhibition of nitrosation, and/or modulation of oncogene or protooncogene expression or function.

Health authorities are actively encouraging the consumption of vegetables through their 5 a day or 7 a day campaigns. This is having an effect on the consumption patterns in developed countries with consumers being more aware of the importance of vegetables in their diets.

There is however a perception that vegetables, and in particular leafy vegetables, can be the source of food poisoning outbreaks. This has in fact been an issue overseas in recent years, with severe outbreaks reported in the USA and Germany due to human pathogen contamination of vegetables. In Australia this has not been an issue, due mostly to the successful implementation of on-farm QA plans that growers must follow before they can sell their products to consumers through markets and supermarkets.

Part of the requirements of a good on-farm QA plan is the sanitary washing of vegetables, especially leafy vegetables, in water containing a sanitiser chemical. This sanitation step will reduce the levels of bacteria, including any contaminating human pathogens, and prevent cross contamination of produce during the washing stage.

In Australia there have been many chemicals available to growers, however growers do not have much independent information available regarding these chemicals.
This project has evaluated a number of chemicals and compared them in terms of efficacy, ease of use, cost and effects on the postharvest shelf life of leafy vegetables.

The results show that Chlorine, as used currently by growers, is still an efficient way to achieve the aims of the sanitation step. An emerging technology using electrified oxidizing water has however shown excellent results and could become a very useful tool for the sanitary washing of vegetables at on-farm level in the near future. The process would fulfil a chemical free washing claim and also be user friendly for the grower; it can be easily adapted to an automatic system with record of efficacy available for review by auditors of on-farm QA systems.

2. Technical Summary

The aim of this project was to compare the efficacy of sanitising chemicals available in Australia in reducing both spoilage and pathogenic microorganisms on vegetables, particularly leafy vegetables. Leafy vegetables are often consumed uncooked and hence the way that they are washed is critical in preventing food safety incidents at point of consumption. The comparison between sanitisers was done in a realistic commercial setting, using equipment already present on farm. The vegetables used in the testing were part of a normal harvest on that same farm. This project

- studied the efficacy of peroxyacetic acid and acetic acid sanitisers

- studied the efficacy of chlorine and chlorobromo sanitisers

- studied the efficacy of organic sanitisers derived from natural material

• studied the efficacy of new and emerging chemical free sanitiser technology

• studied the shelf life of leafy vegetables after treatment with a number of sanitisers.

• extended the knowledge to individual growers on the importance of the sanitation step

• supplied information to the vegetable industry on the correct ways to sanitise leafy vegetables in wash baths and hydro coolers.

• Washing vegetables in water containing 100 ppm of chlorine remains the most suitable system available to growers that wash on-farm at the moment. Chlorine has advantages in terms of efficacy, cost effectiveness and ease of handling. This is closely followed by Chlorobromo sanitisers, which are just as effective but have a slightly higher cost.

3. Key Outcomes and Conclusions

• Growers who produce leafy vegetables that are sold as pre-washed and ready to eat should consider using peroxyacetic acid based sanitisers. These sanitisers are however considerably more expensive and may contribute to a lower shelf life of the product.

• Growers who supply the organic market should consider an organic based sanitiser. Although the efficacy of these is not as good as Chlorine, Chlorobromo, or peroxyacetic acid sanitisers, they still show some level of efficacy and are better than acetic acid.

• In these trials electrified oxidised water, an emerging technology, was shown to have superior efficacy to any of the other products tested. Results also showed that product washed with electrified oxidised water had a longer shelf life.

4. Introduction

Washing of vegetables on-farm is a practice that is used routinely by vegetable growers in Australia. Vegetables need washing for three main reasons: 1) They need washing to remove dirt and dust so that they can be presented to the consumer in a visually appealing manner, 2) They need washing with sanitisers for postharvest treatment purposes so that postharvest plant diseases can be kept under control and thus increase vegetable shelf life and 3) They need washing with sanitisers for food safety purposes so that human pathogens that may be present on the surface of vegetables are not passed on to consumers.

The latter purpose is becoming more and more important as supermarkets, wholesalers and processors are pushing food safety down the line of supply to growers. Vegetable growers however have not kept up with developments in the area of sanitisers and most have not moved away from Chlorine solutions due to the fact that little or no independent information is filtering down to them in terms of new sanitisers. Chemical industry sales representatives are often the only source of information for the growers but this source of advice can be one-sided and skewed towards the product they sell.

In Australia, there have been projects to scientifically evaluate the efficacy of some new sanitisers; some of these have been funded by HAL in the past. The first of these was project VG97045 “New washing treatments for minimally processed vegetables”. This project was carried out by the now defunct AIFSC and was skewed towards the processors of minimally processed vegetables and examined alternatives to Chlorine. One such alternative, which showed a lot of promise, was the use of silver and hydrogen peroxide. This combination showed enhanced bactericidal effect against both spoilage and pathogenic microorganisms in washing trials with fresh-cut produce at bench and pilot scale. However the concept of using silver and hydrogen peroxide on food has not taken off. It has not been seen as a replacement for Chlorine in the fresh-cut industry and has not been extended to washing leafy vegetables on-farm. Since then, many other formats of washing chemicals have been developed but, again, most have not been fully scientifically evaluated. Another project that looked at this was VG99005 “Quality wash water for carrots and other vegetables”. This project, using Ozone and Chlorine dioxide gas, was skewed towards root vegetables, concentrated on re-use of wash water and focused more on the postharvest disease area rather than food safety.

At the present time credible, independent evidence of the efficacy of currently available commercial sanitisers for use predominantly for food safety purposes is lacking in the vegetable industry. More importantly, leafy vegetable growers require this information urgently as there have been a number of overseas food safety outbreaks related to leafy vegetables in the last few years. These outbreaks have led quality assurance system managers to ask what is really known about the efficacy of the current vegetable washing sanitisers in Australia. With the concept of validation now being the buzz word in QA system auditing, the scrutiny of sanitisers will increase.

There are also other pressures resulting in leafy vegetable growers questioning the efficacy of the sanitiser they use, including the fact that there has been a negative response to the use of chlorine in some countries by consumers and regulators. In Australia for example, the organic industry does not allow the use of Chlorine. If organic vegetable growers want to wash leafy vegetables to reduce potential food safety issues, they really do not have credible sources of information to use in setting up a wash system for their products. In addition there is an urgent need to have an emergency plan in place in case Chlorine is banned for use with food in Australia or an export destination.

The sanitation step is becoming more and more important from a food safety perspective. Although there have not been any documented food poisoning outbreaks attributed to leafy vegetables in Australia in the past 10 years, the outbreaks that have occurred overseas have made the news headlines here. These outbreaks include the 2006 *E. coli* outbreak in bagged spinach in the USA, the Romaine lettuce outbreak in the USA in 2011 and the *E.coli* outbreak in Germany in the same year. This has lead to increased scrutiny of the food safety of leafy vegetables by supermarkets, wholesalers and processors within Australia. In addition, there have been numerous human pathogen detections during random testing of leafy vegetables by processors and supermarkets. These detections often lead to suspension of supply from a particular grower until samples show that they are clear of microbial contaminants. This has led to more growers washing leafy vegetables on-farm so as to reduce this risk to their business. There is no doubt that on-farm quality assurance programs such as Freshcare and SQF1000 have reduced this risk to growers, as it is these very programs that have focussed attention on washing systems and sanitisers used in the vegetable industry. As part of the external third party audits, growers need to show that they are maintaining adequate levels of available sanitiser in the wash tanks. This has proven difficult as most growers still use manual dipsticks (based on visual colour checks which are subjective) for testing.

A recent telephone survey conducted as part of this project showed that out of the 30 growers contacted, 27 were using Chlorine, with more than half of these using Calcium Hypochlorite and the rest using Sodium Hypochlorite. These two forms of Chlorination have advantages and disadvantages.

The growers who use Calcium Hypochlorite mostly do this by using tablets that dissolve slowly as water is passed over them. This system can produce huge levels of variability, depending on the flow of the water and on the way the tablets are made. In many cases, this leads to under chlorination, particularly if there is a large organic load (including soil) with the leafy vegetables.

The use of Sodium Hypochlorite on the other hand is easier for some growers as this is available in a liquid form, making it easy for automated dosing systems to add the required volume. An Oxidation Reduction probe can be used to regulate this addition, however these probes are not very accurate and must be checked by hand on a regular basis. This latter step is often overlooked by growers leading to over chlorination resulting in a Chlorine odour and the OH&S issues that are associated with people working in that environment. In addition to this, over-chlorination can lead to rust formation and corrosion of susceptible equipment.

For these reasons, the sanitation step can still be problematic for growers and hence they are continuously seeking a better solution.

5. Review of potential sanitisers to evaluate as part of project

Work by Behrsing *et al* (2000) shows that washing vegetables with potable water has the ability to reduce the levels of bacteria on the surface of the leaf by at least one log. The addition of 100 ppm of Chlorine only serves to further reduce the levels of bacteria on the leaf by less than an additional one log. Therefore the best that can be achieved with Chlorine is a reduction on the leaf of between one and two logs. However, the levels of bacteria in solution in wash water remaining after the washing step when using Chlorine are always close to the levels seen in potable water. The sanitiser then acts in two ways: 1) It further reduces the bacteria on the surface of the leafy vegetable (Beuchat and Ryu, 1997; Sapers, 2001) and 2) it stops cross contamination by killing most of the bacteria that wash off the leaf and into solution.

Both these points are important in deciding which sanitiser to use in the vegetable industry, especially the second point. High loads of pathogens can occur in small segments of the vegetable load being washed. These can occur for various reasons, including localised animal faeces contamination coming in from the field. If the water does not contain an active sanitiser, it is possible that cross contamination of the whole batch can occur. This has occurred in Australia previously when hydro-coolers have been used and the water did not contain an active sanitiser; the water spread the contamination throughout the entire vegetable load.

Currently, the most commonly used sanitiser in the vegetable industry is Chlorine, either in the form of Sodium Hypochlorite ($NaClO$) or Calcium Hypochlorite ($Ca(ClO)_2$). The former is available as a liquid whereas the latter as a solid and is used as a granular additive to a stock solution or as a tablet (Brackett, 1987; Beuchat and Ryu, 1997). Many growers use Sodium Hypochlorite as it is easier to use.

There are however a growing number of alternatives that have recently emerged. Some of these have been used in the food industry for some years but have not been applied as a sanitiser in the fresh produce industry, whereas others have been specifically designed for use in the fresh produce industry.

This review summarises the chemical sanitisers that have been chosen to take part in the evaluation as required by project VG09086. The sanitisers were chosen because 1) they are easily available in Australia, 2) evidence exists that they work as a sanitiser, 3) they have the potential to be used in the vegetable industry and 4) they have the potential to offer some advantage to users in the vegetable industry.

5.1 Peroxyacetic Acid (PAA) containing sanitisers

Peroxyacetic Acid, or Peracetic acid as it is commonly referred to, is manufactured by reacting acetic acid with hydrogen peroxide, resulting in a highly stable PAA solution containing Peracetic acid and hydrogen peroxide. PAA has grown in popularity because of its effectiveness and environmental compatibility. Upon degradation, PAA breaks down into acetic acid (vinegar), water and oxygen.

One of the major advantages in using PAA is that it functions extremely well under cold conditions (4 °C) and unlike other sanitizers, does not experience cold temperature failure. For this reason, sanitization can be carried out on leafy vegetables that have been pre-cooled or in systems that use water to cool the vegetables, such as hydro-coolers.

PAA is registered with FSANZ as a sanitiser for use as a non-rinse washing aid. It is generally used at 50 to 150 ppm and is highly effective against a broad spectrum of bacteria and spores. It has been shown to be particularly effective against *Listeria monocytogenes*; the mechanism is by direct killing of bacteria but it also leaves a residue of acetic acid behind to slow down potential Listeria growth, effectively creating a lag phase for this bacterium on leafy vegetables.

The final major advantage is that it is available in Australia from a range of suppliers and comes with a number of different trade names, including Tsunami, Adoxysan, Summit and Proxitane so that technical support for growers is always available when using these products.

Some disadvantages of PAA are that it is more expensive to apply than hypochlorite and it is difficult to monitor; handheld test strips that show a colour change for PAA are still the best way to measure the levels in solution. Oxidation Reduction probes (ORP) are not sensitive enough to allow for the precise measurement of this oxidiser in solution. Another major disadvantage of this sanitiser is that if it is used at too high a concentration, it can cause damage to the tips of the leaf and it can shorten the shelf life of the produce.

5.2 Stabilised Chlorine Dioxide (ClO_2)

Chlorine Dioxide is usually a preformed gas which is then dispersed into solution. The problem with Chlorine Dioxide gas is that the agitation required to wash leafy vegetables usually causes the gas to come out of solution and dissipate into the atmosphere, potentially causing OH&S issues with operators. Only a few years ago a Chlorine Dioxide gas system installed in a vegetable farm near Melbourne caused the evacuation of the packing shed and the hospitalisation of staff (see appendix 2). However, there is a now a safe alternative in the market as Stabilised Chlorine Dioxide. Stabilized Chlorine Dioxide differs structurally from Chlorine Dioxide gas and is very safe. Stabilized Chlorine Dioxide (SCD or Stabilized ClO_2, also known as Anthium Dioxide and Sodium Chlorite) is a salt form in solution with bicarbonate.

To make Stabilized Chlorine Dioxide, Chlorine Dioxide is taken and manufactured in a liquid state at high pH. Various Sodium Carbonate complexes are then added into the formulation, which link up with and bond to the Chlorine Dioxide to stabilize it. When this is placed in water, the Chlorine Dioxide gas is released. Stabilized Chlorine Dioxide is activated when it encounters bacteria although this is slower than acids and other inorganic substances.

Stabilized Chlorine Dioxide is registered with the US EPA (Environmental Protection Agency) as a bactericide, fungicide and antimicrobial agent. The FDA (Food and Drug Administration) and the USDA (United States Department of Agriculture) have approved the use of Stabilized Chlorine Dioxide in food processing plants for sanitizing and controlling bacteria and mould. In Australia it is registered with FSANZ as a washing aid.

The major advantage of Chlorine Dioxide as a sanitiser are that it is 3 to 4 times as potent as sodium hypochlorite as a sanitising agent and is generally effective against all bacteria and viruses. It does not have the disadvantages that sodium hypochlorite has with respect to corrosion of susceptible metal surfaces. Another advantage is that the stabilised version comes in large tablets for ease of use and from a QA systems point of view it can be controlled and verified with ease by operators. Its main disadvantage is that it may pose OH&S issues if not used correctly and that it may be expensive if not impossible to use on large scale vegetable farms due to the quantity involved.

5.3 Oxidizing water (OW)

The use of OW is another way to inactivate microbes through a form of Chlorination (Izumi, 1999; Sapers, 2001). It differs from commonly used chlorine treatments in that the inactivating agent is generated directly in the water. There are conflicting claims as to where the technology has originated but most probably the technology was originally developed in Russia (Shtannikov et al. 1977).

Oxidizing water does not need special approvals in Australia for direct and indirect food contact applications and can be used as food washing aid without rinsing. Any chemical formed during the electrolysis step reverts back to the same chemicals that existed prior to electrolysis in due time. The process can be termed as chemical neutral as no chemicals are added and none are left behind after processing. The process can sometimes work better when small quantities of salt (NaCl) are added to the wash water but naturally existing salt and salts originating from the cut leaf are often sufficient to form the sanitising reactants.

Oxidizing water is created by the electrochemical disassociation of water and natural salts that may be present in solution as impurities between anode and cathode electrodes in a reaction cell. This process splits water into two separate streams, acidic (anode) and alkaline (cathode) water. OW from the anode stream has at least three antimicrobial properties that include low pH, as low as 2.5, high oxidation-reduction potential and a high level of free chlorine-based compounds (Park *et al.*, 2001). The concentration of the chlorine compounds (usually 20 to 80 ppm) is influenced by the current (amps) of the OW generator. OW also contains a mixture of inorganic oxidants such as HClO, OCl^-, Cl_2, and O_3, which are effective disinfectants (Yang *et al.*, 2003). So the oxidants produced in the process of making OW combine into one effective sanitation step that has the ability to destroy microorganisms in a number of ways thus increasing the sanitation potential.

Oxidizing water has been used for inactivation of a wide variety of pathogenic and spoilage microorganisms, such as *E. coli* (including the O157:H7 strain), *Salmonella enteritidis*, *L. monocytogenes,* and *Campylobacter jejuni* (Park *et al.*, 1999; Venkitanarayanan *et al.*, 1999; Kim *et al.*, 2000; Park *et al.*, 2002; Fabrizio and Cutter, 2003). Several studies have shown that OW is effective at reducing pathogens and/or spoilage microorganisms associated with fresh fruits and vegetables (Izumi, 1999; Kim *et al.*, 2000; Park *et al.*, 2001).

The bactericidal activity of OW is quantitatively correlated to the free chlorine in the form of hypochlorous acid (HOCl) that forms in the solution. On the basis of chemical and spectroscopic data, Nakagawara and others (1998) concluded that the major component of acidic oxidising water is Cl_2/HOCl in chemical equilibrium at given pH values, for example, indications are that the major components are Cl_2 (at pH < 3), HOCl (at pH range 4 – 7), and ClO^- (at pH> 8.5). Some studies have suggested that the very high ORP (oxidation-reduction potential) level of the anode water (> 1,100 mV) is a better indicator of bactericidal activity than pH (Kim *et al.*, 2000; Stevenson *et al.*, 2004). Thus, the concentration of HOCl is correlated to high ORP levels.

The effects of OW were evaluated in a study by Izumi (1999). The study focused on several fresh-cut vegetables, including some of which are normally found in fresh-cut salads (i.e. spinach, sliced carrots and sliced cucumbers). The researcher found that the Total Plate Counts (mesophilic aerobic bacteria) were reduced by 0.6 to 2.6 logs CFU/g when treated with oxidising water containing 15, 30, and 50 ppm available chlorine. The OW containing 50 ppm chlorine had the strongest bactericidal effect. This study noted that the effectiveness of oxidising water was the greatest with leafy vegetables (spinach leaves) which had the maximum surface area/unit weight of tissue among the tested fresh-cut vegetables. The observation that chemical sanitizer's effectiveness is influenced by the type and style of vegetables was also confirmed by Zhang and Faber (1996).

OW has been shown to be effective in reducing human pathogens including *E. coli* O157:H7, *L. monocytogenes*, and *S. Enteritidis*. In a study conducted by Venkitanarayanan *et al* (1999), the effect of OW on the inactivation of *E. coli* O157:H7 and *L. monocytogenes* on the surface of plastic cutting boards was shown. They reported a reduction in the bacterial count > 5.0 log CFU/100 cm^2 and non detectable levels for *E. coli* O157:H7 and *L. monocytogenes* populations on cutting boards, respectively. Their second study (1999), focused on the efficacy of OW for inactivating *E. coli* O157:H7, *S. enteritidis*, and *L. monocytogenes* incubated at different times and temperatures. They reported that *E. coli* O157:H7, *S. enteritidis*, and *L. monocytogenes* were more rapidly inactivated at temperatures greater than 23°C.

A study by Koseki *et al.* (2001), demonstrated that washing lettuce with alkaline OW for 1 minute followed by decontamination with acidic OW water for 1 minute, reduced total aerobic bacteria by 1.7, 1.6, 1.0, and 1.1 logs CFU/g, respectively. Using the same treatment on cabbage, the research reported that total aerobic bacteria were reduced by 1.5, 1.5, 1.5, and 1.0 logs CFU/g, respectively. By contrast, when lettuce was soaked in acidic OW for 10 minutes the viable aerobic bacteria was reduced by 2 logs CFU/g (Koseki *et al.*, 2001). This study concluded that the alkaline OW increased the decontamination of microorganisms on the surface of lettuce leaves. Koseki *et al.* (2004) reported similar results.

Yang *et al.* (2003), reported similar results (2 log CFU/g reduction) when fresh-cut lettuce was treated with 300 ppm of OW at pH 7.0 and 30°C for 5 minutes; these results were also confirmed by Park *et al.* (2001). Swem *et al.* (2002) reported higher *E. coli* O157:H7 reductions (3.0 logs CFU/g) under the same treatment. However there was a difference in the treatments, Swem *et al.* (2002) treated the sample 30 min after inoculation, whereas Yang *et al.* (2003) treated the inoculated lettuce after 24 h of storage at 7°C. Using scanning microscopy, a bacterial film (a sticky and threadlike substance surrounding bacterial cells) was observed on the inoculated lettuce sample after 24 h storage. Therefore, biofilm formation affects the efficacy of OW. Park *et al.* (2001) reported a 2.65 log CFU per lettuce leaf reduction of *L. monocytogenes* inoculated on whole lettuce leaves after 3 minutes treatments. Yang *et al.* (2003) reported a 2.0 and 2.1 log CFU/g reduction when fresh-cut romaine lettuce was treated with 300 ppm OW at pH 7 and 30°C (of *Salmonella* Typhimurium and *L. monocytogenes*, respectively).

Oxidizing water in Australia

Electrified oxidizing water unit manufacturers in Australia are not easy to find. There is however a technology company that has been piloting an OW based unit to treat waste water. The company (Unipolar) claims that their technology (Unipolar® disinfection) is far more advanced than just OW. They claim an additional patent that improves the oxidising potential of their units. Unipolar is a patented electro-chemical technology to generate disinfectants that inactivate pathogens. According to literature supplied by Unipolar it is capable of a more than 4-log inactivation of bacteria and viruses; the technology is designed to take advantage of the compounds inherently present in water to produce a mix of strong oxidising agents, including chlorine, ozone, hydrogen peroxide and hydroxyl radicals. No chemicals are added to produce disinfectants, although the process benefits if there is some salt in the system.

The production of disinfectants at a molecular level and mixing within the cells brings pathogens in intimate contact with disinfectants to maximise their effectiveness.

All water passes through the cells so that all disinfectants formed during the electrolytic reaction start treating the water as soon as they are generated. Additionally, a pulsing current is used for the electrolysis, which is designed to weaken pathogens and make them more susceptible to the disinfectants. There are also significant residual chemicals in the water after treatment. Cl^- for example can be there at over 90ppm, levels sufficiently high to treat the surface of leafy vegetables.

The Unipolar water disinfection technology has been validated in accordance with the Department of Health Victoria guidelines (2010) to inactivate at least 4-log bacteria and viruses in waste water. These guidelines set a higher standard than the US EPA guidelines and 4-log inactivation is the highest credit that can be given to a treatment unit. The unit is claimed by the manufacturers to be safe with no hazardous chlorine or other chemicals added, avoiding the transport, storage and handling of hazardous materials. The system is designed to safely vent hydrogen produced by electrolysis.

One important point is that the whole process can be controlled by automation and the operator interface is simple, making it easy for operators to use the Unipolar equipment. Monitoring and control are based on measurement of residual free chlorine which is easy to record for QA purposes.

It has relatively low power consumption, typically 1.0–1.5 kWh/kL depending on the quality of the water and the level of disinfection required.

Maintenance is low cost and simple, regular electrode cleaning by reverse electrolysis is automated and does not require any chemicals or service water. The short cleaning cycle minimises loss of capacity. Titanium electrodes and PVC cells provide long life and easy maintenance. Individual cells are easily removed.

The modular design of the Unipolar cells allows them to be easily connected in series to enable higher doses of disinfectants in applications where there is a higher water demand. Typically, 4 to 8 Unipolar cells are connected in series to form a module. The capacity of a treatment plant is increased by simply adding more modules in parallel. The arrangement of the cells is compact to minimise the footprint of the installation. This is a system fully supported in Australia by the company with a number of service teams available to assist in the vegetable industry.

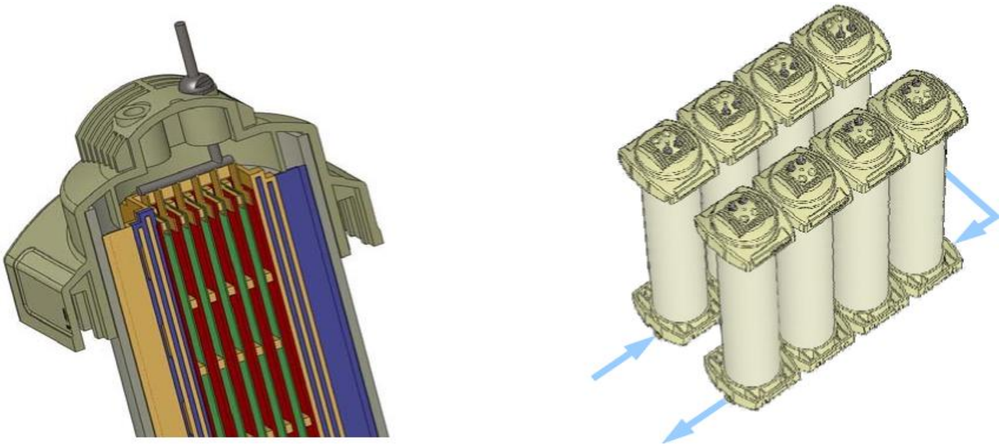

Figure 1. Diagram showing the Electrodes inside a Unipolar cell and a typical module of Unipolar cells (Ref; literature supplied by manufacturer).

5.4 Citrox

Citrox is a formulation of soluble bioflavonoids obtained from citrus fruits. The non-toxic and antimicrobial properties of natural bioflavonoids are well documented, and so these preparations have the potential for application to vegetable washing. This product is recommended by the Australian Organic Council as a washing product for organic vegetables. The core biotechnology of Citrox is the use of complex compounds which have been developed by a team of medical scientists as an antimicrobial for the treatment of oral disease. The key ingredients are flavonoids that are extracted through a specialised process from immature citrus fruits. The extracted flavonoids are combined with a number of natural acids (from fruit and vegetable origins) to produce the core active substances, which have a proven broad spectrum efficacy against harmful micro-organisms, fungi and yeast. A pamphlet published by the makers of Citrox claim inhibition of *E.coli*, *Candita*, *Staphylococcus*, *Salmonella* and *Enterococcus* (http://www.wysonghealth.net/products/documents/monographs/legacy/CITROX.pdf)

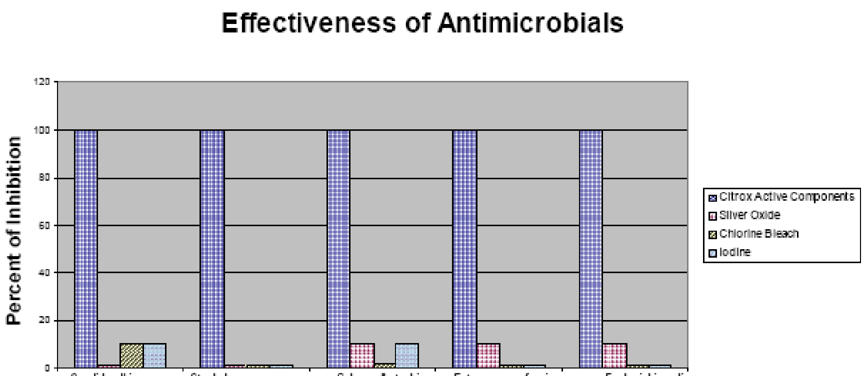

Figure 2; Inhibition of various microbes by the use of Citrox (Ref' http://www.wysonghealth.net/products/documents/monographs/legacy/CITROX.pdf)

Work by S.J. Hooper *et al*. from Cardiff University in the UK has shown that Citrox is an efficient broad-spectrum antimicrobial agent with great potential for use in mouthwashes, playing a role in the maintenance of good oral hygiene.

They have shown that fourteen species of microorganism were selected, including bacteria associated with oral diseases, *Candida* species, and the bacterium *Clostridium difficile*, associated with diarrhoeal infection in hospitalised patients. They used *in vitro* tests to determine the efficacy of two Citrox formulations diluted to 0.007 and 8% v/v. Both Citrox formulations exhibited antimicrobial activity and significantly inhibited growth of all bacteria and yeasts at a working concentration of 1% v/v, however the contact time was not clear.

Citrox has been further developed for use in the washing of fruit and vegetables. Work done by Allende *et al*. (2008) showed that Citrox is just as effective as Chlorine for initial sanitation of leafy vegetables. However, work done by Lopez-Galvez *et al*. (2008) shows that Citrox is perhaps not as efficient as Chlorine in removing cross contaminating *E. coli* from the wash water.

The major advantages of Citrox as stated by the manufacturer are that it is 1) relatively stable at both low and high temperatures, 2) it works best at a pH between 2 and 8 (covering a range of levels found in farm waters), 3) it is manufactured from completely renewable resources, 4) it is effective in the presence of high organic matter loads, 5) it is claimed that it can break up biofilms, 6) it is easy to use and 7) it is completely non toxic and is approved by the Australian Organic Council. The disadvantages include the fact that it can be expensive and it cannot be monitored easily by automatic means (pH readings seem to be the only viable automatic monitoring available).

5.5 Aussan

Aussan is a new generation anti bacterial sanitiser/disinfectant derived from natural and organic ingredients. It is manufactured in Australia from specially selected bioflavonoids and organic acids, which have been derived from vegetables and fruit and give the chemical its efficacy. It is similar to Citrox but does not originate from immature citrus fruit. Aussan is a non toxic organic sanitiser that does not poison the organism but acts mechanistically, by destroying the cellular membrane of a microorganism (cell wall intrusion). All ingredients are carried in a food safe, naturally derived glycerine solvent which makes the active ingredients infinitely soluble in water.

It is a broad spectrum, natural antibacterial sanitiser that can be used in a wide range of industries including, but not limited to, food processing, pharmaceutical, water treatment, catering, medical, agriculture and livestock. When used for sanitising benches, equipment, etc., this powerful agent is claimed by company literature to destroy any bacteria that are present while leaving a non toxic residue that prevents bacteria from multiplying.

Aussan is described as a natural ingredient based sanitizing and preservation agent certified as an input for organic farming. Each ingredient used in the manufacture of Aussan-W4 has been evaluated against the requirements of FSANZ for use in the manufacturing, processing or handling of food. Aussan meets the requirements of FSANZ for use as a processing aid for most raw foods including uncooked crustaceans, fruit and vegetables, and poultry. Where further processing of food occurs such as cooking or preserving, Aussan may also be used as a food additive where required and under GMP, up to a concentration of 5 g/kg Test results supplied by the seller suggest that it can kill 99.86% of a test culture of *Pseudomonas aeruginosa* after a 10 minute exposure. It can also achieve a 99.99 % reduction in *Staphylococcus aureus* after a 10 minute exposure.

Aussan is not toxic to humans, plants or animals. Unlike many other sanitisers, Aussan is not washed from the surface it has sanitised, it remains on the surface and continues to sanitise until the bacteria/pathogen loading exceeds the volume of Aussan present. Other sanitisers and disinfectants must be washed away after use. This means they kill the bacteria, sterilise the surface and are washed away, leaving the surface ready for the next bacteria to arrive.

Advantages of Aussan include that is a wholly natural organic product. It has a broad-spectrum anti-microbial activity, which works against bacteria both gram positive and gram negative. It is non-mutagenic, non-carcinogenic, non-toxic, non-corrosive, non-tainting and non-volatile. It is claimed by the manufacturer to have the ability to break down biofilm and its mechanism of action is by the destruction of the cell wall. It is stable at a wide range of pH levels from 2 to 12 and temperatures of up to 60°C. In fruit and vegetable processing and packing, it can be applied immediately after harvest for protection from human pathogens during picking, handling and transport. According to the manufacturer literature it is an excellent alternative to Chlorine and Peroxy acids in the washing of fruit and vegetables during processing operations as it provides safe and effective residual sanitizing, without the risk of damage to delicate foods and is not deactivated by common soils, so solutions last much longer.

5.6 CitroFresh

CitroFresh is marketed as a food wash and natural preservative and is derived from plant extracts. It can be used as a processing aid and as claimed by sales literature it reduces the risk of food poisoning by eliminating pathogenic micro-organisms (http://www.citrolife.com.au/applications/fruit_and_vegetables). This in turn extends the shelf life and freshness of any fresh, raw, chilled and cooked foods, meats, poultry, fish and produce before packaging and distribution to retail outlets. The manufacturer claims that CitroFresh will keep produce fresh for up to 2-3 times longer than conventional methods, resulting in a very high reduction in spoilage. The product can be applied directly onto food products as a non-rinse sanitiser in either dipping baths, as a fog/mist or a spray. The ten benefits listed in sales brochures include:
1-Organic
2-Non-toxic
3-No harmful chemicals
4-Completely safe, does not invoke pathogen mutations
5-Kills 99.99% of germs
6-Effective against viruses, fungi and bacteria, including *Escherichia coli*, *Clostridium botulinum*, *Listeria monocytogenes*, *Staphylococcus aureus*, *Salmonella choleraesuis*
7-Very effective against biofilms
8-Acts from pH 2 - 12 at temperatures up to 130°C
9-Non-rinse
10-Odourless and colourless: does not change colour or taste of any food
11-Easy to use and store

The major components of this product are citric acid, malic acid, bitter orange extract (bioflavonoid complex) and vegetable glycerine. The product has been shown to be effective at killing *Listeria monocytogenes*, *Salmonella* species and *E. coli* in literature provided by the manufacturer, however when reading this literature carefully the contact times were relatively long for some pathogens. The results of kill rates were also shown when a higher concentration of the product was used at 8%, which is much more than the dose recommended for use in vegetable washing at 2%. The product had many similarities with Aussan.

5.7 Acetic acid

Removal of pathogenic and spoilage bacteria from leafy vegetables by organic acids such as acetic acid have been studied. Acids are either naturally present as constituents of the food or are added to the product through formulation. The undissociated form of acetic acid is utilised to reduce microbial activity, a process which is highly dependable on pH. Nascimento *et al*. (2003) compared the results of sodium hypochlorite with seven different sanitizing solutions (vinegar at 6, 25, and 50 %; acetic acid at 2 and 4 %; peroxyacetic acid at 80 ppm; and sodium dichloroisocyanurate at 200 ppm). The statistical analysis of the results demonstrated that the effectiveness of all the sanitizing agents tested was equivalent to or higher than that for sodium hypochlorite at 200 ppm. The best results were achieved with 4% acetic acid, which reduced the initial aerobic mesophilic population by 3.93 log10 CFU/g and reduced the mould and yeast population by 3.58 log10 CFU/g. Nascimento *et al*. (2003) concluded that the results of the study demonstrated the effectiveness of acetic acid and vinegar as alternative sanitizing agents for the disinfection of fresh produce.

5.8 Nylate

Nylate, also known by the chemical name bromo-chlorodimethyl hydantoin (BCDMH), has been used for over 20 years and has full approvals from both the APVMA and FSANZ for use as a non rinse washing aid in the fruit and vegetable industry. Literature from the distributor states that it forms two extremely effective biocides in solution (hypochlorous acid (HOCI) and hypobromus acid (HOBr)), which have been shown to be effective against fungal and bacterial organisms, as well as a range of human pathogens, such as *Staphylococcus aureus* and *Salmonella kahla*, which pose risks to food safety.

The major advantage of BCDMH is that it is a stable compound, effective across a broad pH range and at much lower concentrations than Chlorine. Whilst Chlorine needs to be used at 100 ppm or more, BCDMH can be used at 5 to 10 ppm. It does however have one disadvantage; when (BCDMH) is hydrolysed, dimethylhydantoin (DMH) is formed as a by-product. Food Standards Australia and New Zealand recommends that the allowable daily intake (ADI) of DMH is 0.025 mg/kg of body weight, which is deemed to be equivalent to concentrations of DMH of 0.2 mg/kg of fruit and 2 mg/kg of vegetables. This always leaves a question mark over the safety of this product, even though it is unlikely that the allowable daily intake limits would ever be reached. The Organic industry has banned the use of BCDMH in washing vegetables.

5.9 Ozone

Ozone is a gas that has to be generated on site as it is very unstable. It is even more unstable in water and over very short periods of time it decomposes into oxygen. At 20 °C it decomposes very quickly with a half life of less than 30 minutes and hence it must be generated continuously (Khadre *et al.*, 2001). In water sources that contain a lot of organic as well as inorganic matter, the activity of ozone will decrease significantly. It is a colourless gas at normal temperatures and is over 3000 times more effective than chlorine oxidising capacity. This means that this sanitiser is very reactive and can corrode metal and other surfaces. It has a pungent smell and is readily detected by the human nose and can cause throat, eye and nose irritation even over short exposures (Guzel-Seydim *et al.*, 2003; Suslow, 1997). It is unlikely that this sanitiser will find extensive use on farms in Australia as it is extremely reactive and poses a significant OH&S risk.

5.10 Concluding commentary;

There are five groups of sanitisers that can be used in the vegetable industry

a) Peroxyacetic acid sanitisers
b) Chlorine sanitisers
c) Organic sanitisers
d) Gas based sanitisers
e) Electrified Oxidizing water

The sanitisers described in this short review all have the potential to be used in the vegetable industry; they also have sufficient scientific evidence that they can actually reduce the levels of bacteria by at least one or two logs. With the exception of electrified oxidising water and Citrox, they are all available commercially at the time of publication of this report. Work done through this project tested all the sanitisers discussed in this report with the exception of Chlorine Dioxide and Ozone. Ozone was not evaluated as part of this project as it needs to be generated on site or purchased as a gas and is difficult to monitor. There was no availability of a processing shed capable of handling Ozone gas based sanitisers at the time of running this project. Chlorine dioxide was also not available for large scale wash tank experiments. A Chlorine dioxide washing system was trialled a number of years ago for washing leafy vegetables. The system failed, gas escaped and the site had to be evacuated and some workers had to be taken to hospital (see appendix 2). As a result it was not evaluated in this study. A tablet that produces chlorine dioxide (Stabilised Chlorine Dioxide) when placed in water is available but it is not feasible in large scale washing systems due to cost. As part of this project, the author has negotiated with Unipolar to develop a small scale electrified water system suitable for evaluation in the vegetable industry.

This study reported on the efficacy of

a) Peroxyacetic acid sanitisers (including Tsunami and Summit)
b) Chlorine sanitisers (Calcium or Sodium Hypochlorite and Nylate)
c) Organic sanitisers (Citrox, Aussan, CitroFresh and acetic acid)
d) Electrified Oxidized water

6. Testing of sanitisers

6.1 General method

All sanitisers were used at the recommended dosage as stated in the accompanying literature or as stated by the supplier; all usage rates are described in the relevant section of the results. Baby spinach was chosen as the leafy vegetable product for the evaluation of the sanitisers as it is intended to be eaten uncooked, is usually very high in microbes and is contaminated with mud and soil. Additionally, most of the sanitisers were also tested with baby leaf lettuce (combination of red/green oak and coral lettuce leaf, rocket and other small leaf) as these are usually sold to be consumed uncooked.

Baths used ranged from 1000 to 2500 litre wash baths, depending on the location used for testing. Two locations were used, both situated on growers' properties on the eastern side of Melbourne and simulated actual washing steps used by the growers. Product was washed as per a commercial batch (usually up to 250 kg of leafy vegetables were washed at any one time in a recirculated wash bath, for the organic based sanitisers and the oxidising water experiments up to 120 kg batches were tested in the smaller wash baths), the contact time of which varied as the product was washed in a turbulent wash bath, but on average the product remained in contact with the sanitiser solution for approximately 45 seconds to two minutes.

Samples of both the product and the wash water were taken after washing produce in clean water (continuous replenishing) and after washing in water containing the sanitiser. These samples were tested in triplicate for total plate counts. Total plate counts or standard plate counts are a useful tool in determining efficacy of sanitisers. Samples were tested according to Australian Standards method (Standard plate count - AS 5013.1-2004, basically Pour plates of appropriate decimal dilutions were prepared using plate count agar and incubated aerobically at 30 ± 1 °C for 72 ± 2 h). Samples of the products were kept for shelf life testing at 4 °C before and after the wash step. *Fluorescent pseudomonads* were also measured but levels were too low to use as indicators for this project. The samples were evaluated for shelf life by visual examination for up to 18 days and checked at days 1, 5, 8, 12, 15 and 18; this was according to the quality scale as shown in appendix1. As a control every treatment was compared to water washing. Water washed produce lasted had a shelf life of 8 days unless otherwise stated.

6.2 Peroxyacetic acid sanitisers

Peroxyacetic acid

Four peroxyacetic acid containing chemicals, that are widely available in Australia, were tested on a washing system typically used on farm to wash produce.

Spinach leaf and baby leaf were washed through the system for a minimum time of 45 seconds (average 60 seconds). Spinach and baby leaf were washed in clean water and two different levels of Peroxyacetic acid sanitiser, namely 40 ppm and 100 ppm. The recommended level is 40 ppm however many users apply it at a higher level of up to 100 ppm.

Microbial counts [Total plate counts (TPC) or standard plate count - AS 5013.1-2004 and expressed as colony forming units (CFU)] were measured in the raw spinach and raw baby leaf before washing and after washing in clean water and water containing sanitiser. Additionally, bacterial counts were also measured in the water and water containing sanitisers after washing. Each test was performed in triplicate and plotted on a bar graph below. The % reduction for each trial of sanitiser compared to water washing was calculated and later plotted on an individual value plot graph. The averages of these three values for each set of trials for each sanitiser was calculated as an average and quoted under the bar graphs below.

The sanitisers tested are shown below, including their composition as declared by the manufacturer:

Summit (Sopura Australia 4 Kingston Park Ct Knoxfield, Victoria, 3180)
Hydrogen peroxide 20%
Peracetic Acid 15%
Acetic Acid 7.5%

Tsunami (Ecolab Australia 6 Hudson Avenue Castle Hill 2154 NSW Australia)
Hydrogen peroxide 7-13%
Peracetic Acid 10-30%%
Acetic acid 15-40%

Adoxysan (Advance Chemicals 4 – 8 Malton Court, Altona Victoria, 3018)
Hydrogen peroxide 10-30%
Peracetic Acid 10 -30%%
Acetic acid less than 10%%

Proxitane (Castle Chemicals Pty. Ltd. 16 Rural Dive Sandgate NSW 2304)
Hydrogen peroxide 20-24%
Peracetic Acid 5-5.4%
Acetic acid 10-12%

Washing with Tsunami.

Spinach washing

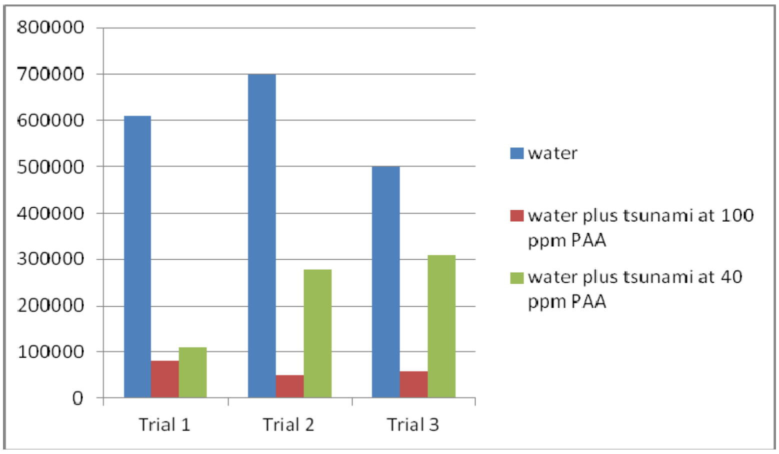

Figure 3; Graph showing the CFU per gram of leaf of spinach in three separate trials using potable water to wash the leaf, potable water plus Tsunami at 100ppm PAA and potable water plus Tsunami at 40 ppm.

Baby leaf washing

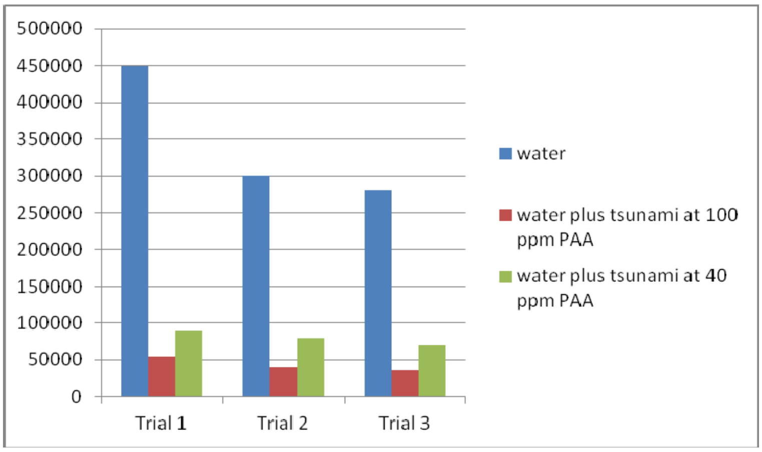

Figure 4; Graph showing the CFU per gram of leaf of baby leaf in three separate trials using potable water to wash the leaf, potable water plus Tsunami at 100ppm PAA and potable water plus tsunami at 40 ppm.

Overall, Tsunami reduced the average levels of total plate counts in spinach by 89.27% (average of three trials) when used at 100ppm PAA and 60.0 % when used at 40ppm PAA when compared to the counts after washing in water. In baby leaf, Tsunami reduced the levels of total plate counts by 87.2% when used at 100ppm PAA and 76.0% when used at 40ppm PAA when compared to the counts after washing in water.

Washing with Adoxysan

Spinach washing

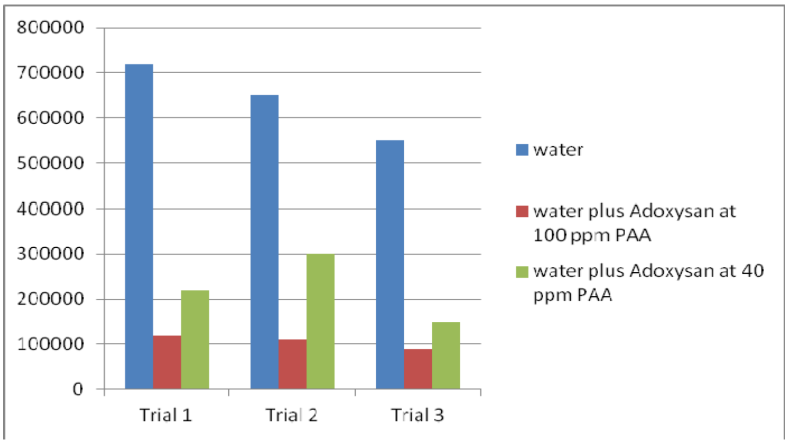

Figure 5; Graph showing the CFU per gram of leaf of spinach in three separate trials using potable water to wash the leaf, potable water plus Anoxysan at 100ppm PAA and potable water plus Anoxysan at 40 ppm.

Baby leaf washing

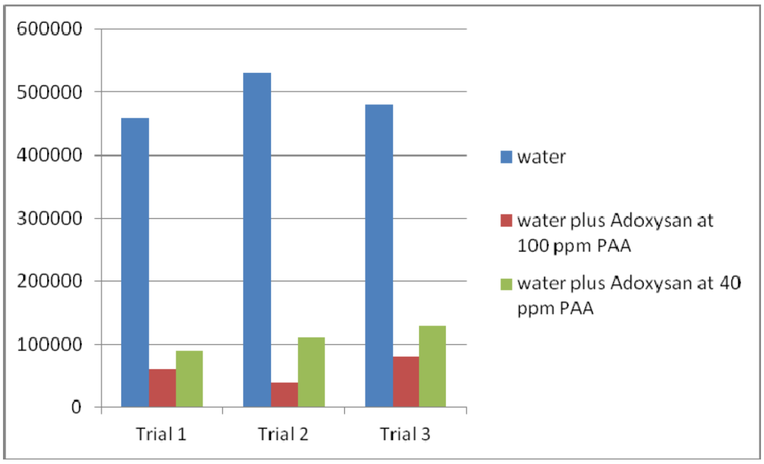

Figure 6; Graph showing the CFU per gram of leaf of baby leaf in three separate trials using potable water to wash the leaf, potable water plus Anoxysan at 100ppm PAA and potable water plus Anoxysan at 40 ppm.

Overall, Adoxysan reduced the average levels of total plate counts in spinach by 83.3% when used at 100ppm PAA and 76.1 % when used at 40ppm PAA when compared to the counts after washing in water. In baby leaf, Adoxysan reduced the levels of total plate counts by 87.8% when used at 100ppm PAA and 77.4% when used at 40ppm PAA when compared to the counts after washing in water.

Washing with Summit

Spinach washing

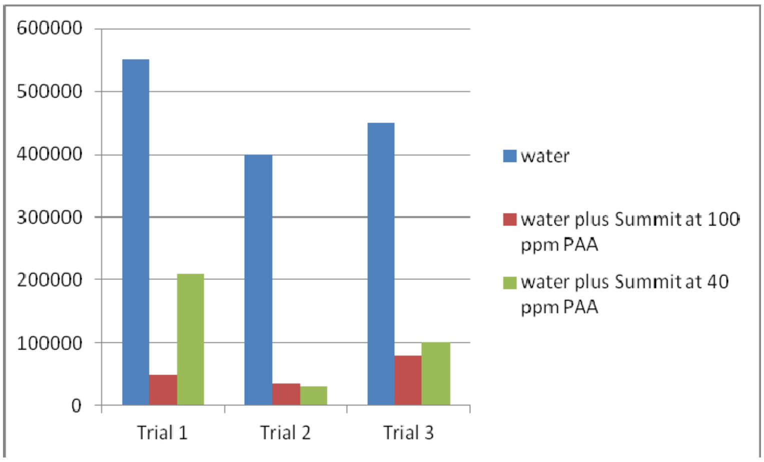

Figure 7; Graph showing the CFU per gram of leaf of spinach in three separate trials using potable water to wash the leaf, potable water plus Summit at 100ppm PAA and poatble water plus Summit at 40 ppm.

Baby leaf washing

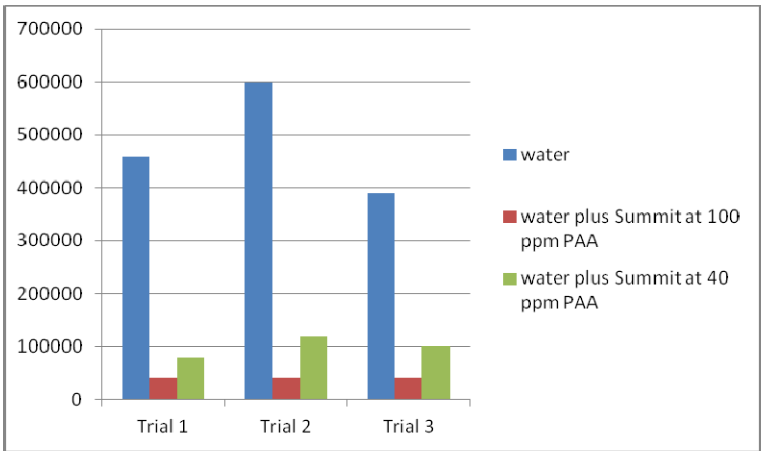

Figure 8; Graph showing the CFU per gram of leaf of baby leaf in three separate trials using potable water to wash the leaf, potable water plus Summit at 100ppm PAA and poatble water plus Summit at 40 ppm.

Overall, Summit reduced the average levels of total plate counts in spinach by 88.1% when used at 100ppm PAA and 72.3 % when used at 40ppm PAA when compared to the counts after washing in water. In baby leaf, Summit reduced the levels of total plate counts by 90.9% when used at 100ppm PAA and 78.3% when used at 40ppm PAA when compared to the counts after washing in water.

Washing with Proxitane

Spinach washing

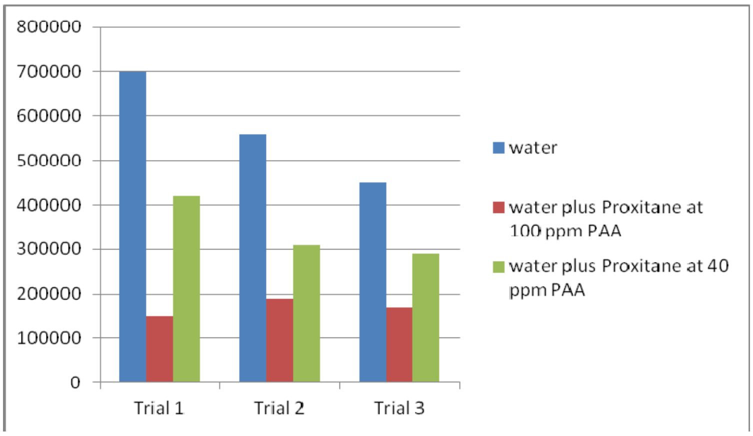

Figure 9; Graph showing the CFU per gram of leaf of spinach in three separate trials using potable water to wash the leaf, potable water plus Proxitane at 100ppm PAA and poatble water plus Proxitane at 40 ppm.

Baby leaf washing

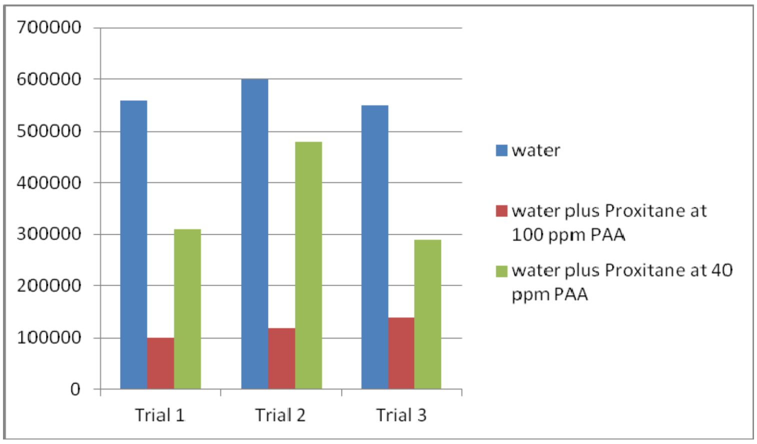

Figure 8; Graph showing the CFU per gram of leaf of baby leaf in three separate trials using potable water to wash the leaf, potable water plus Proxitane at 100ppm PAA and poatble water plus Proxitane at 40 ppm.

Overall, Proxitane reduced the average levels of total plate counts in spinach by 69.0 % when used at 100ppm PAA and 40.1 % when used at 40ppm PAA when compared to the counts after washing in water. In baby leaf, Proxitane reduced the levels of total plate counts to 78.9% when used at 100ppm PAA and 37.3% when used at 40ppm PAA.

Results of shelf life study-

Shelf life of spinach washed in Tsunami at 40ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	4	5	5	5
12	4	4	4	4	4
15	3	3	3	4	3
18	2	2	3	3	3

Shelf life of spinach washed in Tsunami at 100ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	4	5	5	5
8	4	4	4	5	4
12	4	3	4	4	4
15	3	3	3	3	3
18	2	2	3	3	3

Table 1; Table showing the results of the shelf life testing of spinach washed in Tsunami at 40ppm and 100ppm. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 12 when 40 ppm of Tsunami was used and at day 8 when 100 ppm of Tsunami was used.

Shelf life of spinach washed in Adoxysan at 40 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	4	5	5	5
12	4	4	4	4	4
15	3	3	3	3	3
18	2	2	3	3	3

Shelf life of spinach washed in Adoxysan at 100 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	5	4	5	5	5
8	5	4	4	4	5
12	4	3	4	3	4
15	ND	ND	ND	ND	ND
18	2	2	3	3	3

Table 2; Table showing the results of the shelf life testing of spinach washed in Adoxysan at 40ppm and 100ppm. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 12 when 40 ppm of Adoxysan was used and at day 8 when 100 ppm of Adoxysan was used.

Shelf life of spinach washed in Summit at 40 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	5	5	5	5
12	4	4	5	4	5
15	3	3	3	4	3
18	2	2	3	3	3

Shelf life of spinach washed in Summit at 100 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	4	5	5	5
8	5	4	5	4	5
12	4	4	4	4	4
15	3	3	3	4	3
18	2	2	3	3	3

Table 3; Table showing the results of the shelf life testing of spinach washed in Summit at 40ppm and 100ppm. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 12 when 40 ppm of Summit was used and at day 12 when 100 ppm of Summit was used.

Shelf life of spinach washed in Proxitane at 40 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	4	5	5	5
8	4	4	5	5	5
12	4	4	4	4	4
15	3	3	3	4	3
18	2	2	2	3	3

Shelf life of spinach washed in Proxitane at 100 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	4	4	4	4	5
8	4	3	4	4	4
12	3	3	3	3	3
15	3	3	3	3	3
18	2	2	2	3	3

Table 4; Table showing the results of the shelf life testing of spinach washed in Proxitane at 40ppm and 100ppm. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 12 when 40 ppm of Proxitane was used and at day 5 when 100 ppm of Proxitane was used.

Results of shelf life study- Baby leaf

Shelf life of baby leaf washed in Tsunami at 40ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	4	5	5	5
12	4	4	4	4	4
15	3	3	3	4	3
18	2	2	3	3	3

Shelf life of baby leaf washed in Tsunami at 100ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	5	4	5	5	5
8	5	4	5	4	5
12	4	3	4	4	4
15	3	3	3	3	3
18	2	2	3	3	3

Table 5; Table showing the results of the shelf life testing of baby leaf washed in Tsunami at 40ppm and 100ppm. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 12 when 40 ppm of Tsunami was used and at day 8 when 100 ppm of Tsunami was used.

Shelf life of baby leaf washed in Adoxysan at 40 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	5	4	5	5	5
8	5	4	5	4	5
12	4	4	4	4	4
15	3	3	4	3	3
18	2	2	3	3	3

Shelf life of baby leaf washed in Adoxysan at 100 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	5	4	5	5	5
8	4	4	4	5	5
12	4	3	4	4	4
15	ND	ND	ND	ND	ND
18	2	2	3	3	3

Table 6; Table showing the results of the shelf life testing of baby leaf washed in Adoxysan at 40ppm and 100ppm. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 12 when 40 ppm of Adoxysan was used and at day 8 when 100 ppm of Adoxysan was used (ND signifies that no data is available for those days).

Shelf life of baby leaf washed in Summit at 40 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	4	4	5	5	5
12	4	4	4	4	4
15	3	3	3	4	3
18	2	2	3	3	3

Shelf life of baby leaf washed in Summit at 100 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	5	4	5	5	5
8	4	4	5	5	5
12	3	3	4	4	4
15	3	3	3	3	3
18	ND	ND	ND	ND	ND

Table 7; Table showing the results of the shelf life testing of baby leaf washed in Summit at 40ppm and 100ppm. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 12 when 40 ppm of Summit was used and at day 8 when 100 ppm of Summit was used (ND signifies that no data is available for those days).

Shelf life of baby leaf washed in Proxitane at 40 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	4	4	5	5	5
8	4	3	4	4	4
12	3	3	4	3	4
15	2	2	3	3	3
18	2	2	2	3	3

Shelf life of baby leaf washed in Proxitane at 100 ppm

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	4	4	4	4	5
8	3	3	4	3	4
12	3	4	4	3	3
15	2	2	3	3	3
18	ND	ND	ND	ND	ND

Table 8; Table showing the results of the shelf life testing of baby leaf washed in Proxitane at 40ppm and 100ppm. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 8 when 40 ppm of Proxitane was used and at day 5 when 100 ppm of Proxitane was used (ND signifies that no data is available for those days).

The shelf life studies showed that there were notable differences between the washing solutions. Product washed in Proxitane showed a slightly reduced shelf life when the product was used at 100 ppm. This could be due to the fact that it contains half the level of PAA as compared to other products evaluated and therefore, to get the required PAA level into the final wash, this means also increasing the level of hydrogen peroxide in the final solution. Hydrogen peroxide is known to have harmful effects on leaf at higher levels (Upadhyaya et al 2007). Where ND appears samples went missing and no data was available.

Level of TPC in the wash water

Good sanitisers should be able to kill all bacteria in the wash water, irrespective of how dirty the vegetable is. In this part of the experiment, water samples were taken after washing the produce and the TPC was measured to indicate the level of bacteria that remained behind after washing relatively dirty spinach. The water colour was dark brown suggesting that a lot of soil and dust had been extracted during the washing procedure. Water on its own and water plus 40 ppm PAA and 80 ppm PAA for each of the four PAA based sanitisers after washing the spinach was utilised to measure the TPC.

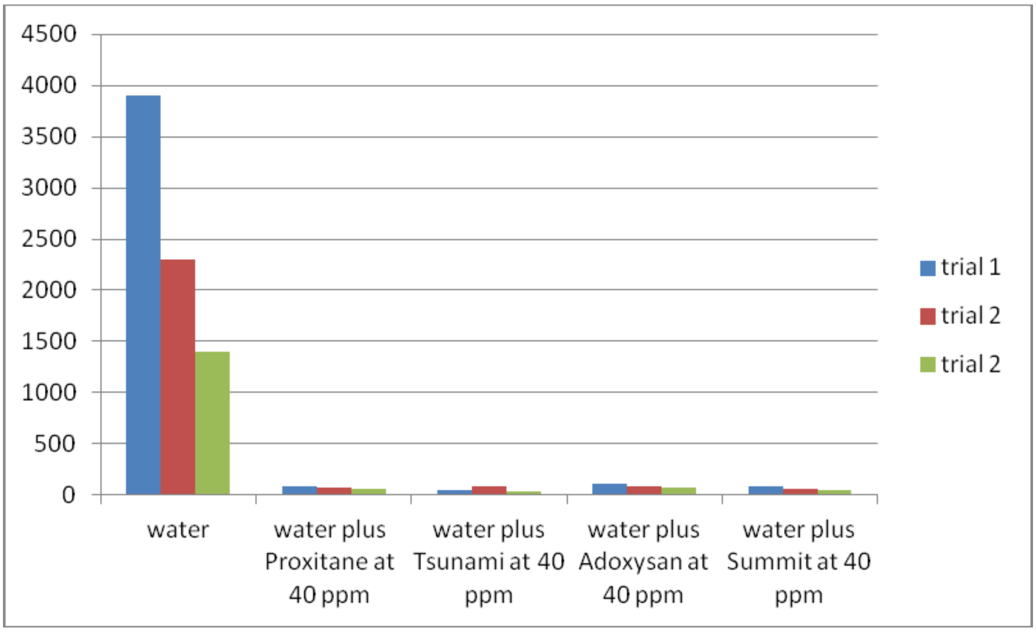

Figure 9; Graph showing the CFU per ml remaining in the wash water after washing spinach. The levels left behind were all below 105 CFU/ml and were similar for each sanitiser used.

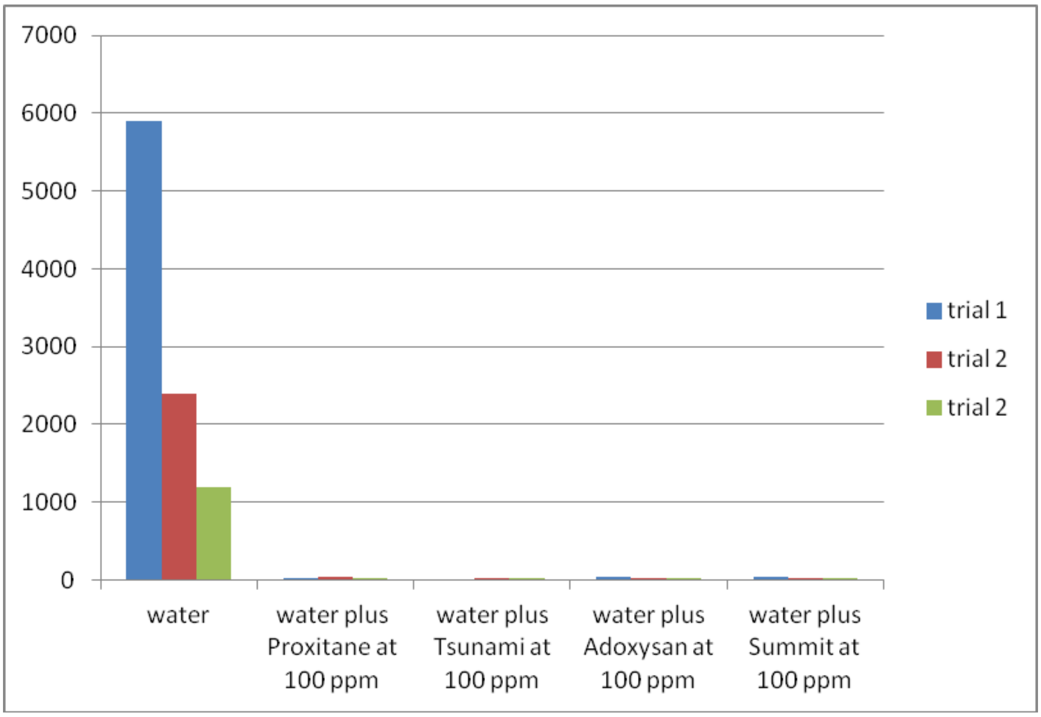

Figure 10; Graph showing the CFU per ml remaining in the wash water after washing of baby leaf. The levels left behind were all below 45 CFU/ml and were similar for each sanitiser used.

The results show that relatively high level counts could be detected in the water after it was used to wash the spinach. The bacteria that are washed off the spinach remain viable in the water and actually build up in numbers as more of the product is washed. Any human pathogens surviving the sanitation step would be able to cross contaminate other produce that is introduced into the wash water. The levels in the same water containing 40 ppm of PAA showed very low levels of TPC and even lower when the PAA was set at 100 ppm. This suggests that the sanitisers have good sanitising properties in the wash water and spinach washed in this sanitiser would unlikely be cross contaminated by the wash water. Results for wash water after washing baby leaf were similar.

6.3 Acetic acid

Acetic acid (Sigma chemicals) was tested on a washing system typically used on farm to wash produce at two concentrations, 2%v/v and 4% v/v in water.

Spinach leaf and baby leaf were washed through the system for a minimum time of 45 seconds (average 60 seconds). Spinach and baby leaf were washed in clean water and the two different levels of Acetic acid.

Microbial counts [Total plate counts (TPC) or Standard plate count - AS 5013.1-2004 as measured in colony forming units (CFU)] were measured in the raw spinach and raw baby leaf before washing and after washing in clean water and water containing sanitiser. Additionally, bacterial counts were also measured in the water and water containing sanitisers after washing. Each test was performed in triplicate and plotted on a bar graph below. The % reduction for each trial of sanitiser compared to water washing was calculated and later plotted on an individual value plot graph. The averages of these three values for each set of trials for Acetic acid was calculated as an average and quoted under the bar graphs below.

Spinach washing

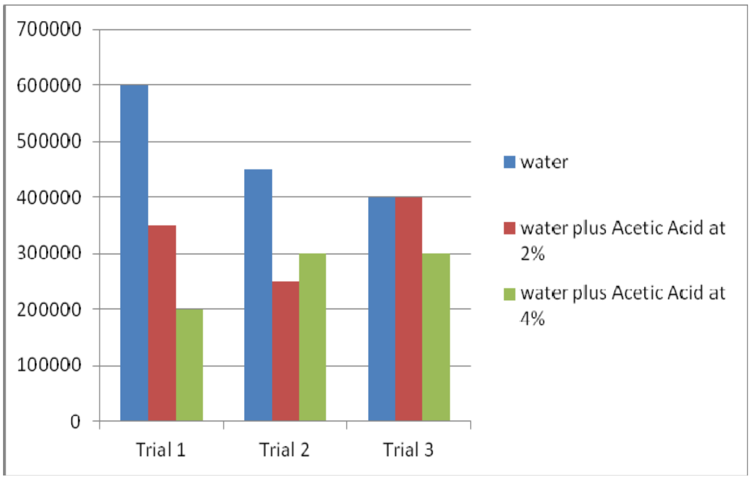

Figure 11; Graph showing the CFU per gram of leaf of spinach in three separate trials using potable water to wash the leaf, potable water plus 2% Acetic acid and potable water plus 4% Acetic acid.

Baby leaf washing

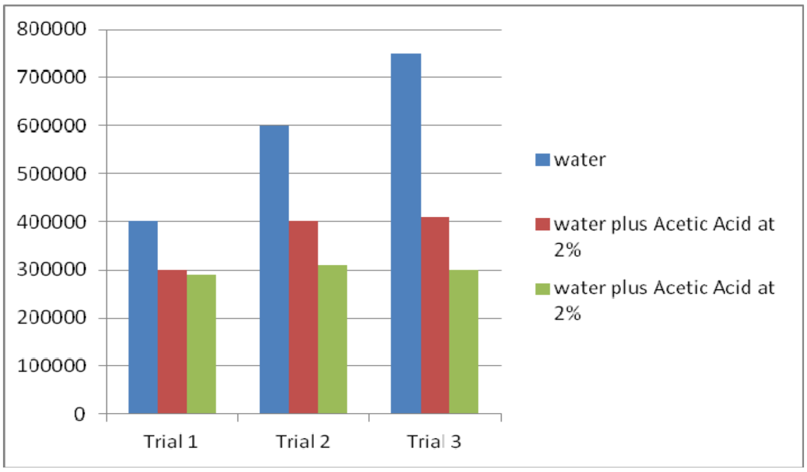

Figure 12; Graph showing the CFU per gram of leaf of baby leaf in three separate trials using potable water to wash the leaf, potable water plus 2% Acetic acid and potable water plus 4% Acetic acid.

Overall, Acetic acid reduced the average levels of total plate counts in spinach by 45.5% when used at a concentration of 4% v/v and 28.7 % when used at a concentration of 2% v/v when compared to the counts after washing in water. In baby leaf, Acetic acid reduced the levels of total plate counts by 45.3 % when used at a concentration of 4% and by 34.5% when used at a concentration of 2%.

Shelf life study

Shelf life of spinach washed in Acetic acid at 2% v/v

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	4	5	5	5
8	4	4	4	4	4
12	3	3	4	4	4
15	3	3	3	3	3
18	2	2	3	2	3

Shelf life of spinach washed in Acetic acid at 4% v/v

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	5	4	5	5	5
8	4	4	4	4	4
12	3	3	4	3	4
15	3	3	3	3	3
18	2	2	2	2	3

Table 9; Table showing the results of the shelf life testing of spinach washed in 2% and 4% Acetic acid. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 8 when 2% Acetic acid was used and at day 8 when 4% Acetic acid was used.

Shelf life of baby leaf washed in Acetic acid at 2% v/v

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	4	4	5	5	5
8	4	4	4	4	4
12	3	3	4	3	4
15	3	3	3	3	3
18	2	2	2	3	3

Shelf life of baby leaf washed in Acetic acid at 4% v/v

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	4	4	5	5	5
8	3	3	4	4	4
12	3	3	4	3	4
15	3	3	3	3	3
18	2	2	2	3	3

Table 10; Table showing the results of the shelf life testing of baby leaf washed in 2% and 4% Acetic acid. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 8 when 2% Acetic acid was used and at day 5 when 4% Acetic acid was used.

The shelf life results clearly show that the higher concentrations of Acetic acid affected the shelf life of the leaf. For baby leaf washed in 4% v/v acetic acid the shelf life was reduced substantially from product washed in water alone which averaged a shelf life of 8 days.

Level of TPC in the wash water

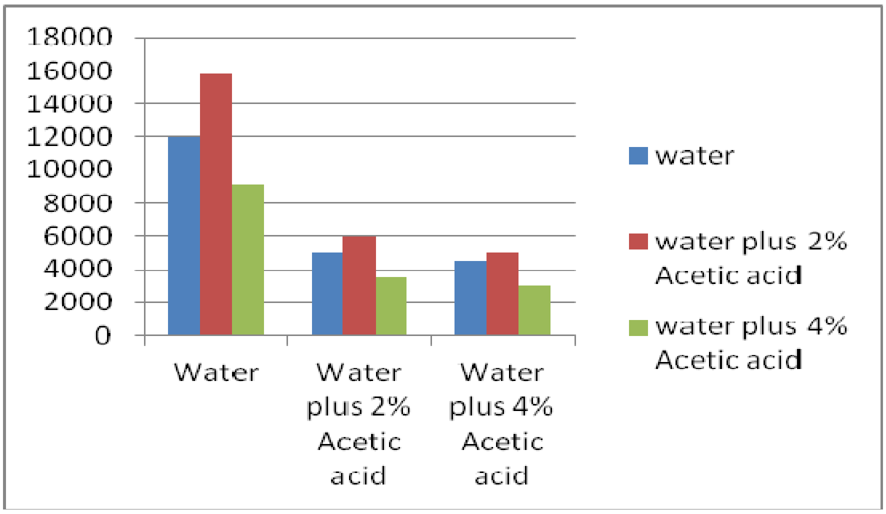

Figure 13; Graph showing the CFU per ml left in the wash in three separate trials using potable water to wash the leaf, potable water plus 2% Acetic acid and potable water plus 4% Acetic acid.

The levels of TPC in the water were only reduced by half in the presence of 2% or 4% acetic acid. This suggests that acetic acid does not have a satisfactory sanitising action to kill any bacteria that is washed from the leaf.

6.4 Organic Acid based sanitisers

Citrox

Citrox was used at two concentrations, 1% and 2.5% v/v in water and spinach leaf was washed through the system for a minimum time of 45 seconds (average 60 seconds). The wash tank used was unmodified and was part of a standard wash system as used on a vegetable farm. Product was tested before treatment, treatment in clean water and treatment in water with the sanitiser added. Microbial counts were measured in the raw spinach before washing and after washing in each of the above solutions. Additionally, bacterial counts were also measured in the different wash waters used. Each test was performed in triplicate and graphed. Shelf life was tested over 18 days post washing.

Spinach washing

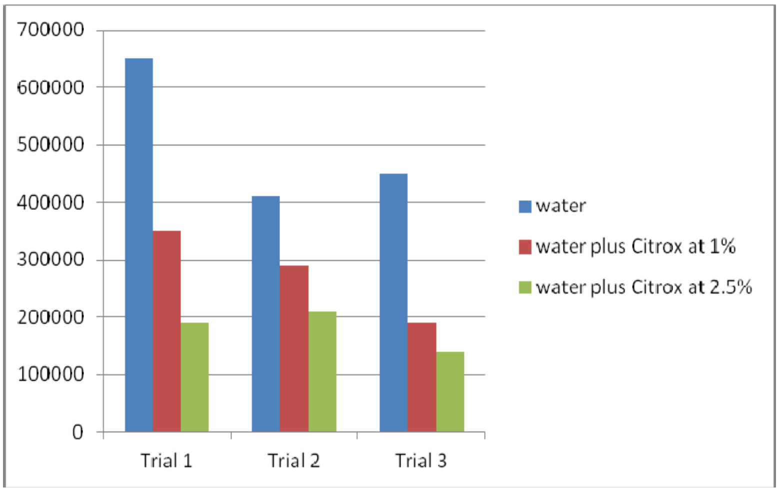

Figure 14; Graph showing the CFU per gram of leaf of spinach in three separate trials using potable water to wash the leaf, potable water plus 1% Citrox and potable water plus 2.5% Citrox.

Baby Leaf washing

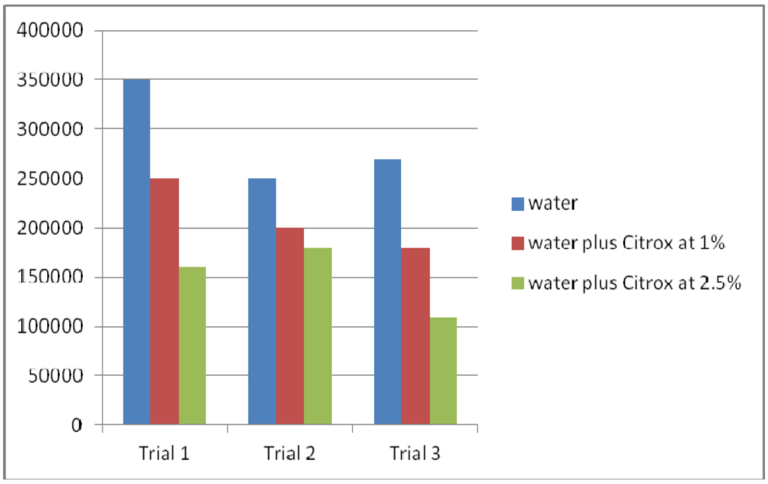

Figure 15; Graph showing the CFU per gram of leaf of baby leaf in three separate trials using potable water to wash the leaf, potable water plus 1% Citrox and potable water plus 2.5% Citrox.

Overall, Citrox reduced the average levels of total plate counts in spinach by 62.8% when used at a concentration of 2.5% v/v and by 44.4 % when used at a concentration of 1% v/v when compared to the counts after washing in water. In baby leaf, Citrox reduced the levels of total plate counts by 47.2% when used at a concentration of 2.5% v/v and by 27.3% when used at a concentration of 1% v/v.

Shelf life study

Shelf life of spinach washed in 1% v/v Citrox

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	4	4	5	4	4
12	3	3	4	3	3
15	2	2	3	3	3
18	2	2	3	2	3

Shelf life of spinach washed in 2.5% v/v Citrox

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	4	4	5	5	5
8	3	3	4	4	4
12	2	2	2	3	3
15	2	2	2	2	3
18	2	2	2	2	2

Table 11; Table showing the results of the shelf life testing of spinach washed in 1% and 2.5% Citrox. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 8 when 1% Citrox was used and at day 5 when 2.5% Citrox was used.

Shelf life of baby leaf washed in 1% v/v Citrox

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	4	4	5	5	5
8	4	4	5	4	4
12	3	3	4	3	3
15	2	2	3	3	3
18	2	2	3	2	3

Shelf life of baby leaf washed in 2.5% v/v Citrox

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	4	4	4	5	4
8	3	3	4	4	4
12	2	2	3	3	3
15	2	2	2	2	3
18	1	2	1	1	2

Table 12; Table showing the results of the shelf life testing of baby leaf washed in 1% and 2.5% Citrox. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 8 when 1% Citrox was used and at day 5 when 2.5% Citrox was used.

The results show that the higher levels of Citrox decreased the shelf life of the produce to 5 days, which is 3 days less than for produce washed in normal water. Commercial use of the product would have to be justified on grounds other than shelf life.

Levels of TPC in the water

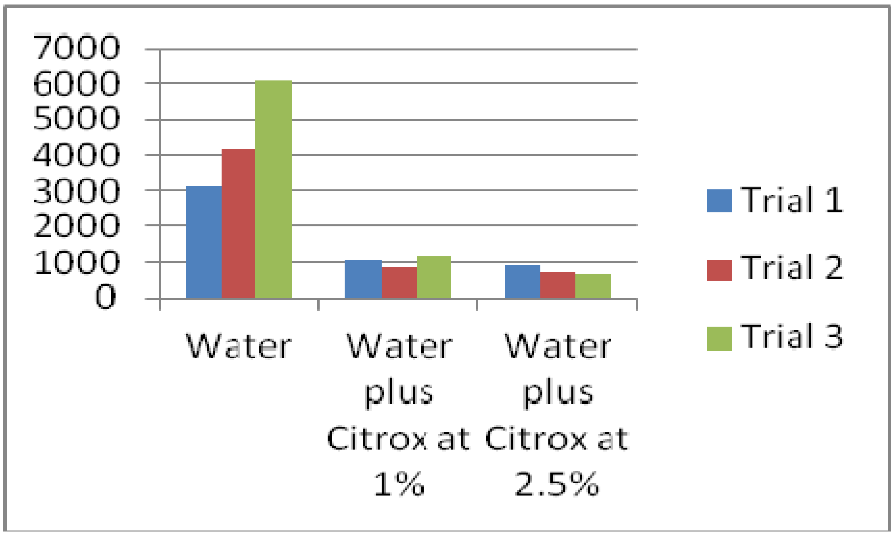

Figure 16; Graph showing the CFU per ml left in the wash in three separate trials using potable water to wash the leaf, potable water plus 1% Citrox and potable water plus 2.5% Citrox.

The graph shows water that was tested on its own, after it had been used to wash spinach, and also a Citrox solution of 1% and 2.5% v/v. There were substantial numbers of bacteria left behind in the wash water after washing spinach. The levels were relatively high, suggesting that as a sanitiser, Citrox does not kill all the bacteria in the water. Results for baby leaf were similar.

Aussan

Aussan was used at two concentrations, 1% and 2.5% v/v in water and spinach leaf was washed through the system for a time of 45 seconds (average 60 seconds). Product was tested before treatment, after treatment in clean water and after treatment in water with the sanitiser added at the two concentrations. Microbial counts were measured in the raw spinach before washing and after washing in each of the above solutions. Additionally, bacterial counts were also measured in the water and water containing sanitisers after washing. Each test was performed in triplicate and graphed. Shelf life was tested over 18 days.

Spinach washing

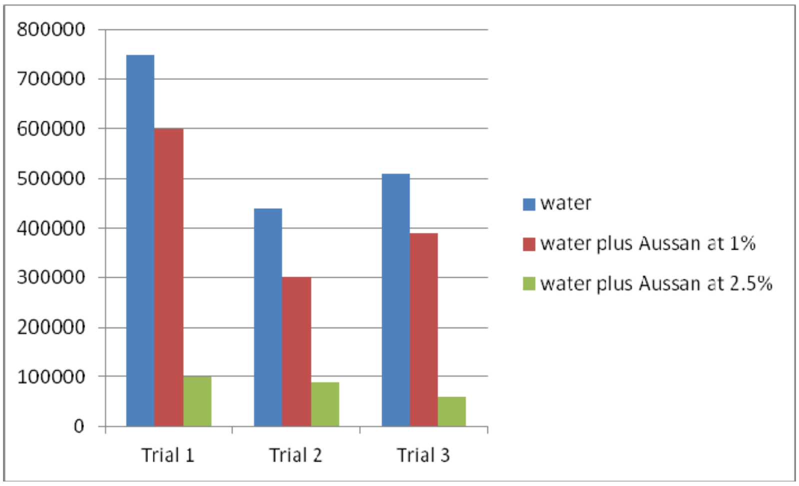

Figure 17; Graph showing the CFU per gram of leaf of spinach in three separate trials using potable water to wash the leaf, potable water plus 1% Aussan and potable water plus 2.5% Aussan.

Baby leaf washing

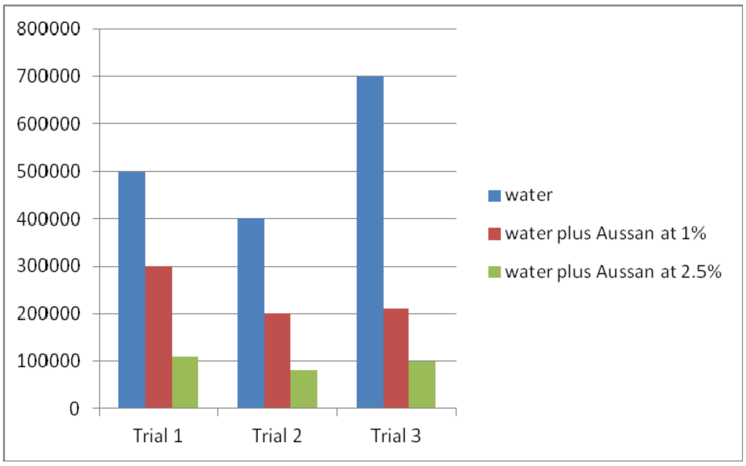

Figure 18; Graph showing the CFU per gram of leaf of baby leaf in three separate trials using potable water to wash the leaf, potable water plus 1% Aussan and potable water plus 2.5% Aussan.

Overall, Aussan reduced the average levels of total plate counts in spinach by 84.8% when used at a concentration of 2.5% v/v and by 25.1 % when used at a concentration of 1%. In baby leaf Aussan reduced the levels of total plate counts by 81.2% when used at a concentration of 2.5% v/v and by 53.3% when used at a concentration of 1% v/v.

Shelf life study

Shelf life of spinach washed in 1% v/v Aussan

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	5	5	5	5
12	4	4	5	5	5
15	3	3	4	4	4
18	2	2	3	2	3

Shelf life of spinach washed in 2.5% v/vAussan

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	4	4	4	4	4
12	3	4	4	3	4
15	2	3	3	2	3
18	2	2	2	2	2

Table 13; Table showing the results of the shelf life testing of spinach washed in 1% and 2.5% Aussan. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 12 when 1% Aussan was used and at day 8 when 2.5% Aussan was used.

Shelf life of baby leaf washed in 1% v/v Aussan

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	5	5	5	5
12	3	3	4	3	4
15	2	2	3	3	3
18	2	2	3	2	3

Shelf life of baby leaf washed in 2.5% v/v Aussan

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	4	4	4	5	4
8	4	4	4	4	4
12	3	2	3	3	3
15	2	2	2	2	3
18	2	2	2	2	2

Table 14; Table showing the results of the shelf life testing of baby leaf washed in 1% and 2.5% Aussan. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 8 when 1% Aussan was used and at day 8 when 2.5% Aussan was used.

The results show that at both levels of Aussan, the shelf life of the produce was similar to that obtained with produce washed in untreated water (8 Days). There is some evidence that when used at 1% v/v Aussan increases the shelf life to 12 days.

Levels of TPC in the water

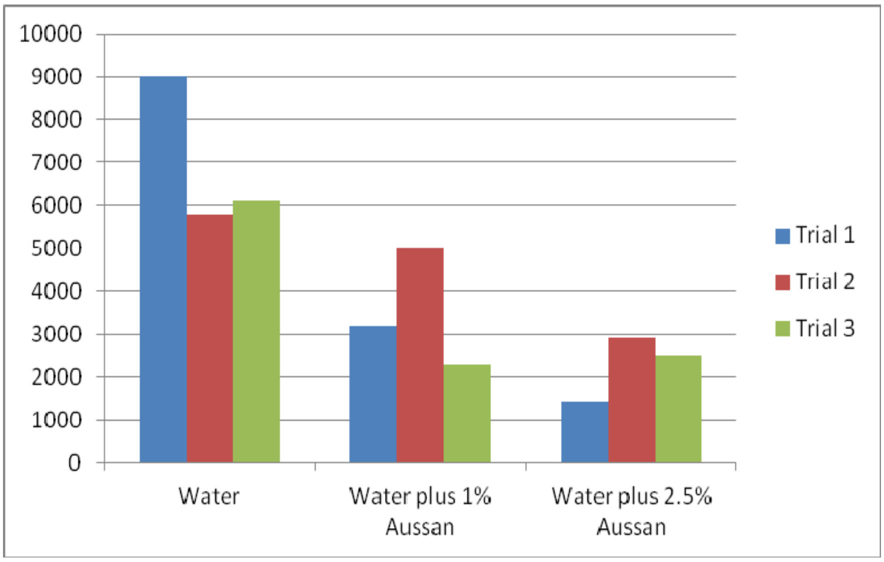

Figure 19; Graph showing the CFU per ml left in the wash in three separate trials using potable water to wash the leaf, potable water plus 1% Aussan and potable water plus 2.5% Aussan.

This graph shows water tested for TPC on its own after it had been used to wash spinach and also for a 1% and 2.5% v/v Aussan solution. There were substantial numbers of bacteria remaining in the wash water after washing the spinach. The levels were relatively high suggesting that as a sanitiser, Aussan does not effectively kill bacteria in the water. Results for baby leaf were similar and are not shown here.

CitroFresh

Two Citrofresh solutions were prepared at a concentration of 1 % and 2.5% v/v. These 2 solutions, together with untreated water, were used to wash spinach and baby leaf separately through the system for a time of 2 to 4 minutes (average 2.5 minutes). Product was tested before treatment, treatment in clean water and treatment in water with the sanitiser added. Microbial counts were measured in the raw spinach before washing and after washing in each of the above solutions. Additionally, bacterial counts were also measured in the water and water containing sanitiser after washing. Each test was performed in triplicate and graphed. Shelf life was tested over 18 days.

Spinach washing

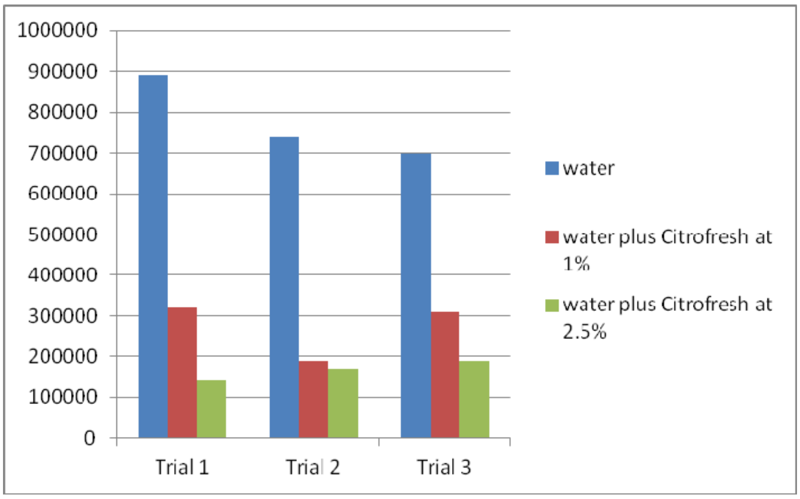

Figure 20; Graph showing the CFU per gram of leaf of spinach in three separate trials using potable water to wash the leaf, potable water plus 1% CitroFresh and potable water plus 2.5% CitroFresh.

Baby leaf washing

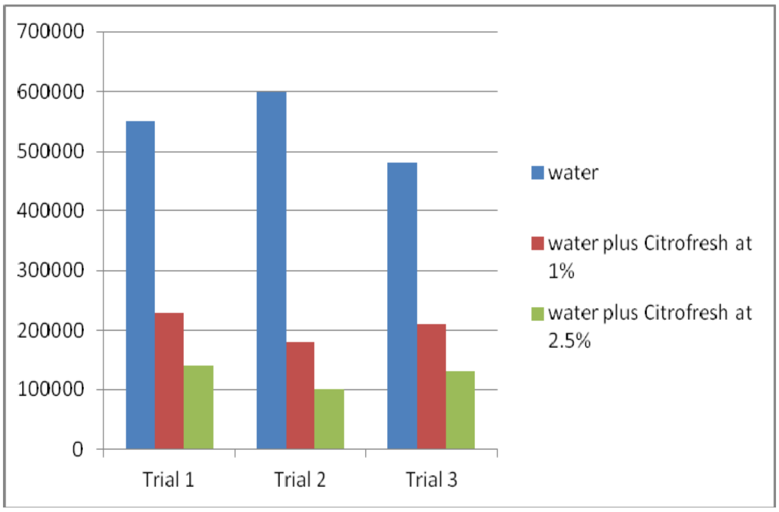

Figure 21; Graph showing the CFU per gram of leaf of baby leaf in three separate trials using potable water to wash the leaf, potable water plus 1% CitroFresh and potable water plus 2.5% CitroFresh.

Overall, CitroFresh reduced the average levels of total plate count in spinach by 78.1% when used at a concentration of 2.5%v/v and by 64.7 % when used at a concentration of 1% v/v as compared to the counts after washing in water. In baby leaf, CitroFresh reduced the levels of total plate counts by 76.9% when used at a concentration of 2.5% v/v and 61.5% when used at a concentration of 1% v/v.

Shelf life testing

Shelf life of spinach washed in 1% v/v CitroFresh

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	5	5	5	5
12	4	4	5	5	5
15	3	3	4	4	4
18	2	2	3	2	3

Shelf life of spinach washed in 2.5% v/v CitroFresh

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	4	4	4	4	4
12	4	4	4	4	4
15	2	3	3	3	3
18	2	2	2	2	2

Table 15; Table showing the results of the shelf life testing of spinach washed in 1% and 2.5% CitroFresh. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 12 when 1% CitroFresh was used and at day 12 when 2.5% CitroFresh was used.

Shelf life of baby leaf washed in 1% v/v CitroFresh

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	4	5	4	5	5
12	4	4	4	4	4
15	3	3	3	3	3
18	2	2	3	2	3

Shelf life of baby leaf washed in 2.5% v/v CitroFresh

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	4	5	4	5	4
8	4	4	4	4	4
12	3	4	4	3	4
15	2	2	2	2	3
18	2	2	2	2	2

Table 16; Table showing the results of the shelf life testing of baby leaf washed in 1% and 2.5% CitroFresh. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 8 when 1% CitroFresh was used and at day 8 when 2.5% CitroFresh was used.

CitroFresh achieved a relatively good shelf life of 12 days for spinach at both concentrations, for baby leaf however it was only able to achieve a shelf life of 8 days.

Levels of TPC in the water

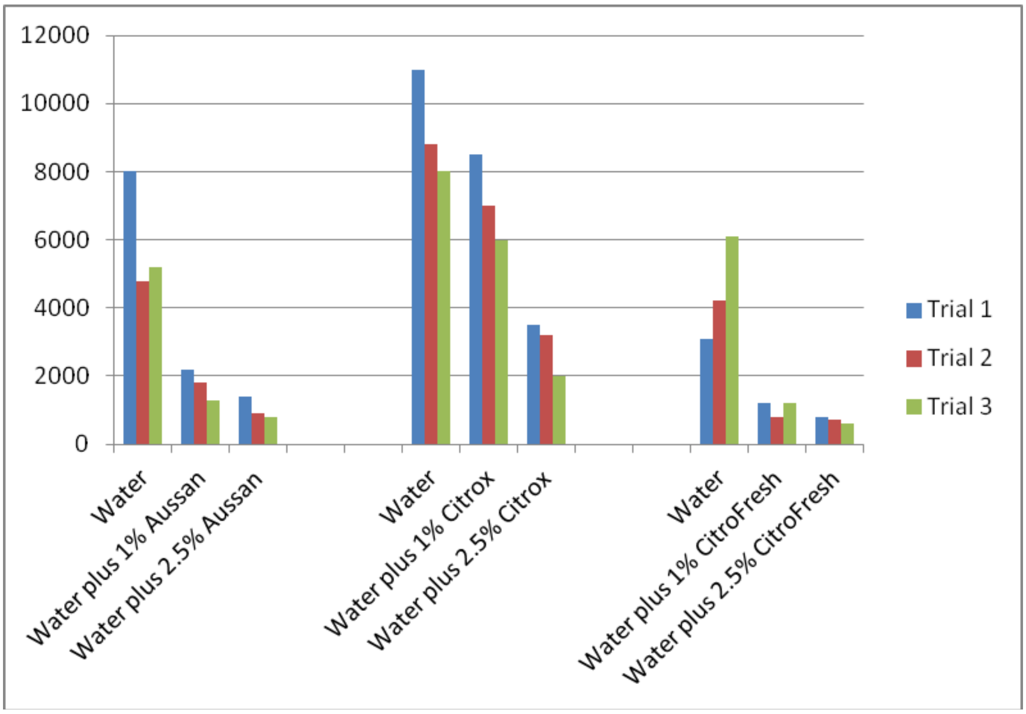

Figure 22; Graph showing the CFU per ml left in the wash in three separate trials using potable water to wash the leaf, potable water plus 1% Aussan, Citrox or CitroFresh and potable water plus 2.5% Aussan, Citrox or CitroFresh

The results shown in figure 22 clearly show that TPC are not eliminated in the wash water after washing in the presence of 1% and 2.5% Aussan. The levels, however, were lower in water containing 2.5% of CitroFresh or Aussan as compared to 2.5% Citrox.

6.5 Chlorine and Nylate based sanitisers

Chlorine in the form of Sodium hypochlorite (Sigma Chemicals) and Nylate (Wobelea, 18 Embrey Court, Pakenham, Victoria 3810) were tested on a washing system typically used on farm to wash produce. Chlorine was used at 100 ppm free chlorine and Nylate was used at 5 ppm. Concentrations were checked by dipsticks analysis every 15 minutes during the trials. Spinach leaf and baby leaf were washed through the system for a minimum time of 45 seconds (average 60 seconds). Spinach and baby leaf were washed in clean water and in Chlorine at 100 ppm and Nylate at 5 ppm.

Chlorine (Sodium Hypochlorite)

Spinach washing

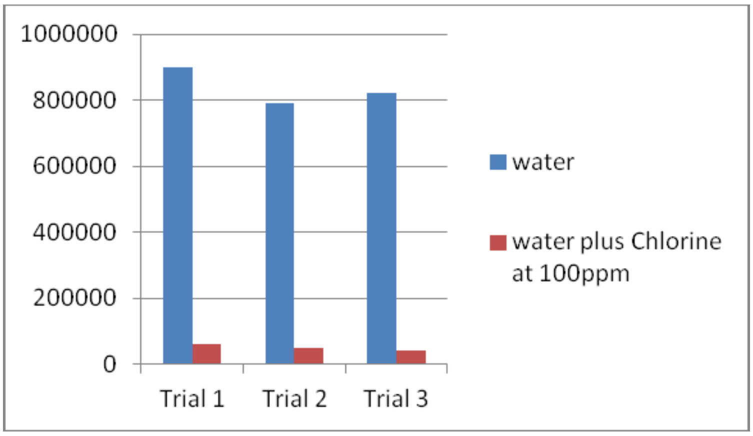

Figure 23; Graph showing the CFU per gram of leaf of spinach after using potable water to wash the leaf and potable water plus 100ppm of Chlorine in three trials.

Overall, 100 ppm of available Chlorine in the washing solution reduced the levels of total plate count by 89.3%

Baby leaf washing

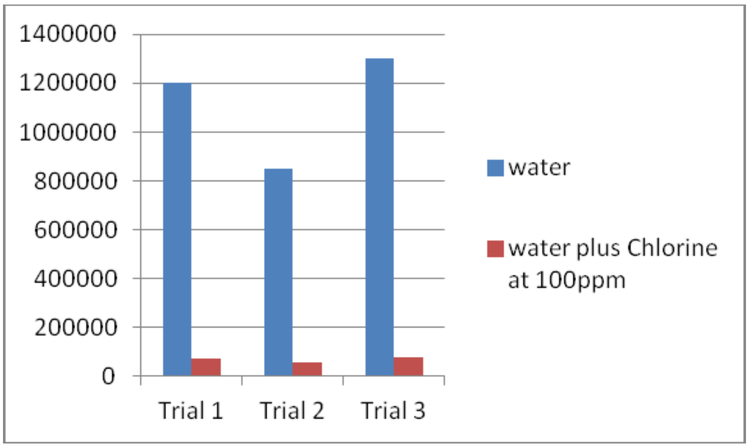

Figure 23; Graph showing the CFU per gram of leaf of baby leaf after using potable water to wash the leaf and potable water plus 100ppm of Chlorine in three trials.

Overall, Chlorine reduced the average levels of total plate count in spinach by 93.5% when used at a concentration of 100ppm as compared to the counts after washing in water. In baby leaf, Chlorine reduced the levels of total plate count by 94.0% when used at a concentration of 100ppm.

Shelf life testing

Shelf life of spinach washed in 100ppm of Chlorine

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	4	5	4	5
12	4	4	4	4	5
15	3	3	3	4	4
18	2	2	2	3	3

Shelf life of baby leaf washed in 100ppm of Chlorine

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	4	5	5	5
12	4	4	4	4	5
15	3	3	3	4	4
18	2	2	2	3	3

Table 17; Table showing the results of the shelf life testing of spinach and baby leaf washed in100 ppm of Chlorine. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 12 for both spinach and baby leaf when 100 ppm of Chlorine was used.

The results show that the shelf life obtained after washing in 100 ppm of Chlorine can be up to 12 days; this is perhaps the benchmark in terms of shelf life, as it is what is currently used in the industry by many processors.

Nylate

Spinach washing

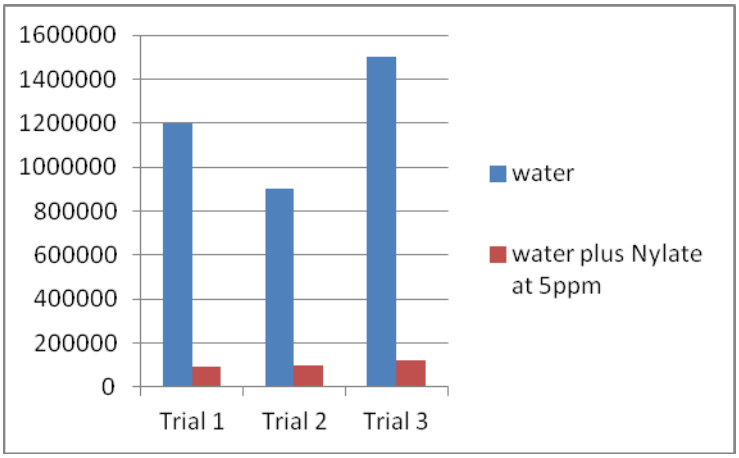

Figure 24; Graph showing the CFU per gram of leaf of spinach after using potable water to wash the leaf and potable water plus 5ppm of Nylate in three trials.

Baby leaf washing

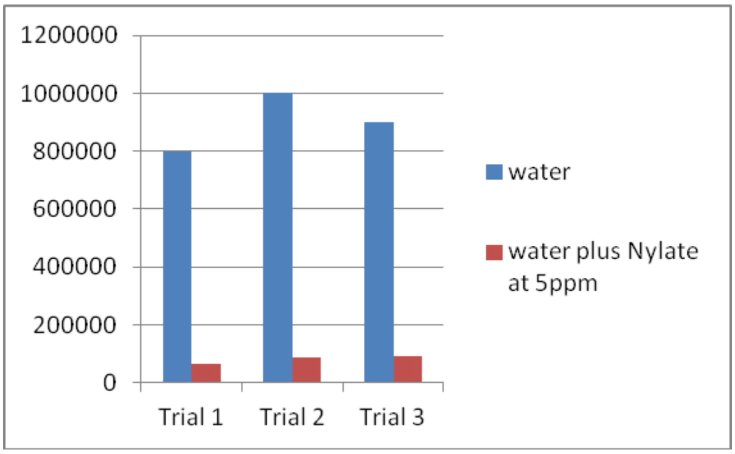

Figure 25; Graph showing the CFU per gram of leaf of baby leaf after using potable water to wash the leaf and potable water plus 5ppm of Nylate in three trials.

Overall, Nylate reduced the average levels of total plate count in spinach by 91.1% when used at a concentration of 5ppm as compared to the counts after washing in water; in baby leaf, Nylate reduced the levels of total plate count by 91.3% when used at the same concentration.

Shelf life testing

Spinach after washing in 5ppm Nylate solution

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	4	5	4	5
12	4	4	4	4	5
15	3	3	3	4	3
18	2	2	2	3	3

Baby leaf after washing in 5ppm Nylate solution

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	4	5	5	5
5	5	4	5	5	5
8	5	4	5	5	5
12	4	4	4	4	5
15	3	3	3	4	3
18	2	2	2	3	3

Table 18; Table showing the results of the shelf life testing of spinach and baby leaf washed in5 ppm of Nylate. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 12 for both spinach and baby leaf when 5 ppm of Nylate was used.

The results shown in table 18 suggest that the shelf life obtained after washing in 5 ppm of Nylate can be up to 12 days, which matches the benchmark standard of 100 ppm of Chlorine.

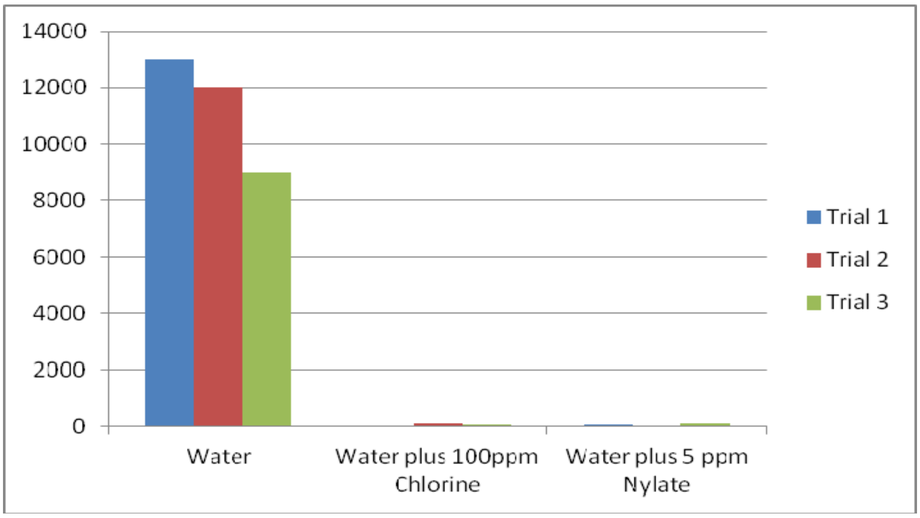

Figure 26; Graph showing the CFU per ml left in the wash in three separate trials using potable water to wash the leaf, potable water plus 100 ppm of Chlorine and potable water and 5 ppm of Nylate.

The results shown in figure 26 show that after washing spinach, water containing either 100ppm of Chlorine or 5 ppm of Nylate both contain very low levels of TPC, no level was higher than 40 CFU/ml. Results for water remaining after washing baby leaf contained similar levels of TPC.

6.6 Oxidising water Unipolar® Disinfection

The Unipolar® disinfection technology is designed to generate biocides directly from the wash water, including chlorine, hydrogen peroxide and ozone. To test the Unipolar® system as a potential sanitiser for use on farms, a mobile Unipolar® unit was connected to a typical wash tank in a vegetable farm with a primary and a secondary washing tank. Spinach was washed at three different power settings; each was designed to deliver a specific level of biocides produced by electrolysis of the water.

Each setting was monitored by measuring the residual free chlorine produced in the electrolytic cells. Although a number of chemical moieties with capacity to sanitise produce can be generated, chlorine is the most stable to measure and thus was used as a guide to control the Unipolar® system. The three power settings were designed to deliver approximately 20 ppm, 45 ppm and 60 ppm of residual free chlorine in the wash water. The product (spinach and baby leaf) was washed through the system for a minimum time of 45 seconds (average 60 seconds). Product was tested after washing in clean water and after washing in water disinfected by the Unipolar® system. Microbial counts were measured in the raw spinach and raw baby leaf before washing and after washing in each of the above solutions. Additionally, TPC was measured in the untreated water and in the Unipolar® disinfected water after washing. Each test was performed in triplicate and graphed. Shelf life was tested over 18 days.

Spinach washing with Unipolar® disinfected water

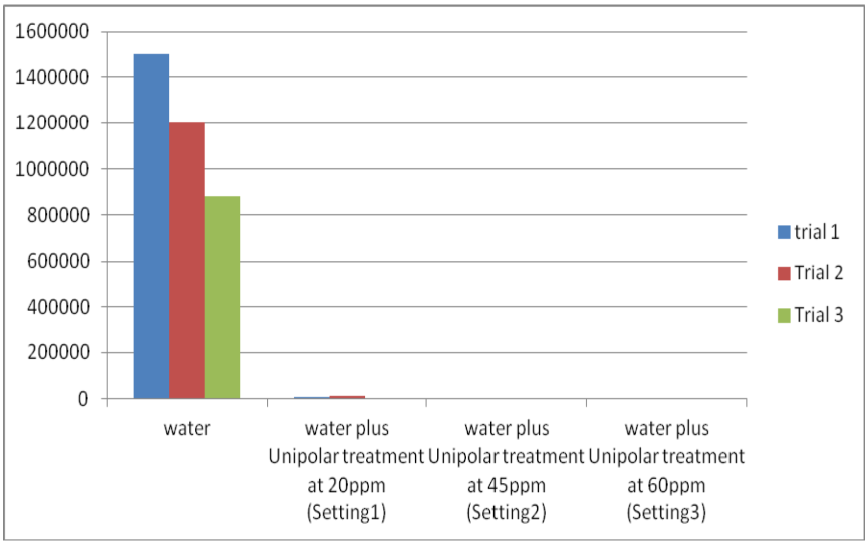

Figure 27; Graph showing the CFU per ml left in the wash in three separate trials using potable water to wash the leaf, potable water plus Unipolar treatment at 20ppm (Setting 1), 45ppm (Setting 2) and 60 ppm (Setting 3).

Overall, Unipolar® disinfected water reduced the levels of total plate count by 99.6% with setting one, 99.9% with setting 2 and 3.

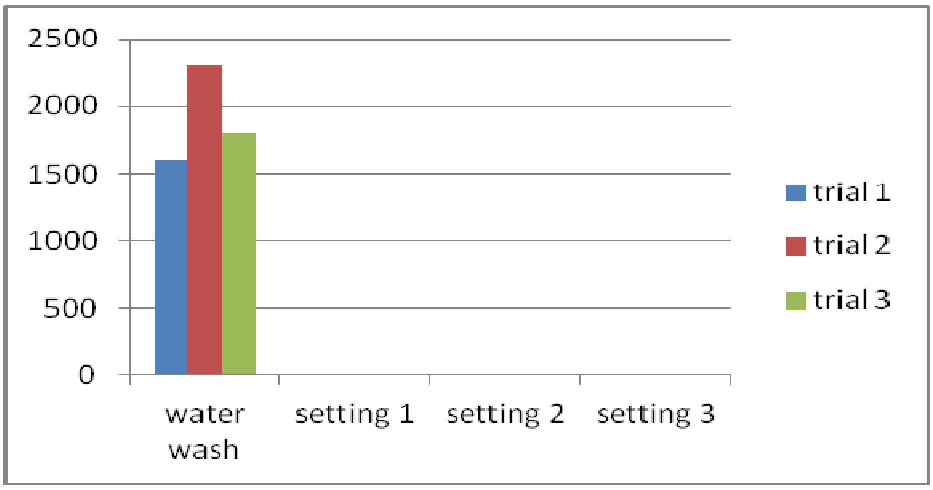

Figure 28; Graph showing the CFU per ml left in the wash in three separate trials using potable water to wash the leaf, potable water three settings of Unipolar disinfected water corresponding to 20ppm, 45ppm and 60ppm of disinfectant.

The results shown in figure 28 show the levels of bacteria left in the water after washing spinach; the actual levels in Unipolar® disinfected water were all below 2 CFU/ml for the 3 different power settings used.

Shelf life

Shelf life of spinach washed in Unipolar® disinfected water (level1)

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	5	5	5	5
12	5	5	5	5	5
15	4	4	4	4	4
18	3	3	3	3	3

Shelf life of spinach washed in Unipolar® disinfected water (level 2)

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	5	5	5	5
12	5	5	5	5	5
15	4	4	4	4	5
18	3	3	3	3	3

Shelf life of spinach washed in Unipolar® disinfected water (level 3)

Day	OVQ	Wetness	Discolouration	Wilting	Rots
1	5	5	5	5	5
5	5	5	5	5	5
8	5	5	5	5	5
12	4	5	5	5	5
15	3	3	3	4	5
18	3	3	3	3	4

Table 19; Table showing the results of the shelf life testing of spinach washed at three levels of Unipolar disinfection. The results show that the minimum score of 4 required for full consumer acceptance was reached at day 15 for both spinach when setting 1 was used (20ppm of Unipolar disinfection), day15 when setting 2 was used (45ppm of Unipolar disinfection) and day 12 when setting 3 was used (60ppm of Unipolar disinfection),

The results shown in table 19 suggest that at settings 1 and setting 2 of Unipolar disinfection spinach showed a shelf life of 15 days, exceeding that of Chlorine. At setting 3of Unipolar disinfection, the shelf life was the same as that obtained with the benchmark standard of Chlorine.

7. Discussion of results

The results for all sanitisers and treatments were analysed by the statistical unit at the University of Melbourne. The variation in TPC levels on the leaf makes it impossible to do a direct comparison between products as these were treated on different days. Different sites and produce were also obtained from different fields at different times of the year. The only tool available to do a comparison is to use the level of TPC in produce after washing in water and compare that to the level of TPC after washing in a sanitiser solution. A percentage change can then be calculated between the 2 treatments, using water as the control. The results were then able to be graphed as individual value plots, which could then be compared to gauge how the sanitisers performed.

Plot 1 shows the individual value plots of the percentage change in the baby leaf TPC between water and sanitiser washing. The results show that only four sanitisers managed to exceed the 90 % mark: these included Nylate at 5ppm, Chlorine at 100ppm, Summit at 100 ppm PAA, and some of the Adoxysan treated products. Very close to the 90% mark was Tsunami at 100 ppm PAA. A number of sanitisers did not make it to the 40% mark. These included Acetic acid at both concentrations tested, Citrox at 1% and some of the 2.5% Citrox treatments and one of the Proxitane treatments. Aussan was just on the border of the 40% mark. Overall, the best sanitisers were Chlorine and Nylate with the peroxyacetic acid sanitisers following closely behind. The organic sanitisers did not perform as well as the above mentioned sanitisers.

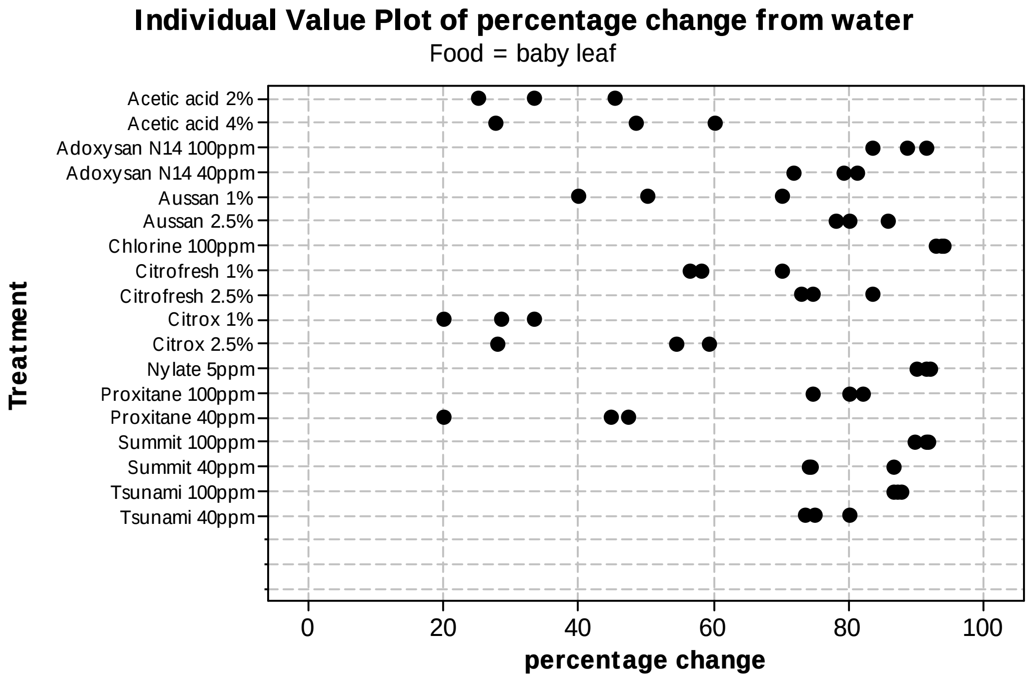

Figure 29; Graph of individual value plot (Plot 1) of % change from water for baby leaf (Unipolar disinfection not shown)

Results for washing spinach are shown in Plot 2. The individual value plots of the percentage change in the spinach TPC between water and sanitiser washing show that five sanitiser treatments managed to exceed the 90% mark. These include some of the Aussan at 2.5% v/v, Chlorine at 100ppm, Nylate at 5ppm, Summit at 100 ppm, and Tsunami at 100ppm. In this plot, the results for the Unipolar disinfected water are also shown. These are very close to the 100% mark, making them the most efficient sanitising treatment studied in this report, even exceeding that of Chlorine at 100 ppm. A number of sanitiser treatments did not meet the 40% mark, which included Citrox at 1% v/v, some of the Proxitane treatments at 40ppm PAA, Aussan at 1% v/v, both Acetic acid treatments and Tsunami at 40ppm PAA came close to the 40% mark.

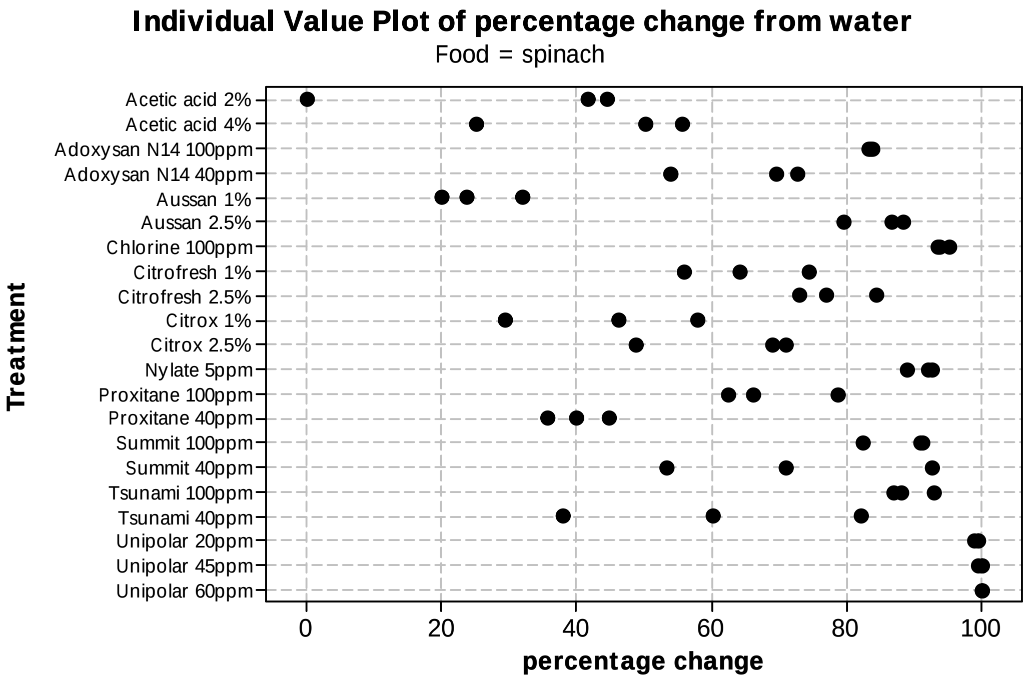

Figure 30; Graph of individual value plot (Plot 2) of % change from water for spinach

Washing in plain water alone gives a product which has a shelf life of 10 days for Spinach and 8 days for baby leaf (results not presented here but conducted as part of the shelf life trial). Discussions with growers suggest that there is an expectation that baby leaf and spinach reach a minimum shelf life of 12 days, something that is achievable with Chlorine washing. The sanitation step is also seen as a way to increase the shelf life of produce. Not all of the sanitising treatments however achieved this expectation, many only reached 8 days and one only managed to reach 5 days. The Peroxyacetic acid sanitisers mostly achieved 12 days shelf life when treated with 40 ppm of sanitiser. The shelf life decreased slightly to 8 days as the level of PAA sanitiser increased to 100ppm, with the exception of Proxitane, which only achieved a shelf life of 5 days when used at 100ppm PAA. Acetic acid at both the concentrations used in this study achieved a shelf life of 8 days with the exception of baby leaf treated with 4% v/v Acetic acid that only reached a 5 day shelf life. The organic sanitisers showed a variable shelf life: Citrox showed an 8 day shelf life when used at 1% v/v but the shelf life was reduced to 5 days when it was used at 2.5% v/v; Aussan showed a longer shelf life reaching 8 days even when used at 2.5% v/v.

CitroFresh showed the longest shelf life of 12 days when used at 1% v/v for baby leaf but with spinach, this shelf life was also achieved when used at both 1% and 2.5% v/v.

Chlorine and Nylate also showed a shelf life of 12 days but the real surprise was the shelf life observed with the Unipolar sanitised water, showing a shelf life of up to 15 days with treatments at settings one and two.

8. General Discussion

The levels of TPC in leafy vegetables can be extremely variable and can change from field to field and from planting to planting. However, CFU's are often used by most auditors of a HACCP plan to verify the sanitation step. The auditors look for a two log reduction in bacteria after the sanitation step. This has been relatively hard to achieve in all cases so auditors accept a one log reduction as evidence that the sanitation step has worked. In reality, what the auditors should also be looking for is the elimination of bacteria left behind in the wash water.

There is no doubt that the levels left behind in the wash water are relatively low when the sanitiser used is Chlorine, Nylate or the Peracetic acid sanitisers. Acetic acid, on the other hand, does not effectively reduce bacteria in the wash water. The use of acetic acid is not wide spread in the vegetable industry. A small number of organic growers however have used it or are still using it. The results presented here show that Acetic acid when used at 2% and 4% v/v has little effect on the levels of bacteria on the surface of spinach and baby leaf. At 2% and 4% v/v the levels on the surfaces of the leaf were reduced at best by approximately 50%. This level of reduction would not satisfy current QA requirements for food safety. The levels of bacteria remaining in the wash water were also relatively high, again suggesting that Acetic acid does not meet the requirements of a sanitiser. The shelf life of the finished product was also compromised when using Acetic acid at both concentrations trialled. Spinach had no problems meeting an 8 day shelf life when washed at both concentrations. However, baby leaf only achieved a 5 day shelf life when it was washed with the 4% v/v solution of Acetic acid. With the 2% v/v solution it reached an 8 day shelf life.

The Peroxyacetic acid sanitisers all showed acceptable results, however it is important to note that there is a difference in the formulations of these products. This difference can have an effect on the efficacy of the sanitisers and on the shelf life of the product being sanitised. The level of peroxide is very different in Proxitane when compared to the other preparations. In addition to this, the level of Peroxyacetic acid is also much lower in this commercial preparation meaning that more of the solution needs to be used to achieve the desired PAA concentration in the wash bath. As a result of this, the level of peroxide in the wash bath increases thus affecting the shelf life of the product being sanitised. Overall however, the use of Peroxyacetic acid for washing leafy vegetables was shown to be satisfactory in terms of efficacy and shelf life. Their use in a recirculating wash bath with contact times of less than two minutes was shown to be satisfactory in a commercial situation. Continuous checking needs to take place by using test strips that measure PAA, something that can be expensive.

The organic sanitisers did not perform as well as some of the other sanitisers tested. These sanitisers are often used to wash produce destined for the organic market. The commercial use of these sanitisers is however restricted to small scale production. A disadvantage of these sanitisers is that their efficacy is not as good in killing bacteria when compared to some of the other sanitisers tested in this report, especially in solution and on the leaf. All the literature examined by the suppliers of these organic sanitisers make claims about 99.9% killing capacity against a number of pathogenic bacteria. It is only when one dwells on the conditions of the test that long contact times are noted. Some of these contact times exceed 45 minutes, something that cannot be achieved in a commercial situation when contact times of less than two minutes are essential. Another major disadvantage of these sanitisers is the fact that they cannot be used easily on a recirculating water bath as they have a tendency to foam. Additionally, checking for levels of sanitiser is extremely hard and they are more suitable for a batch washing system, making their use expensive.

The most suitable sanitisers still appear to be the Chlorine or Nylate based sanitisers. Both work in separate manners but have extremely good efficacy in wash baths. The chlorine system needs to be pH monitored and should not exceed a pH of 7; for the trials conducted in this report, this was achieved with citric acid. Failure to adjust the pH of the wash bath could mean an ineffective sanitation step. Nylate does not need to be pH adjusted. Both sanitisers can be monitored in the wash baths with the use of test strips or Oxidation/Reduction probes.

The shelf life obtained when using these sanitisers was higher in most cases than that obtained when using other sanitisers. The major disadvantage of using Chlorine is perhaps the threat that Chlorine may be banned from food production as it has been in a number of countries, especially in Europe. Chlorine may react with organic material to form undesirable by-products such as Chloroforms and other tri-halomethanes, which may have carcinogenic potential. At high pH, chlorine reacts with organic nitrogen based material to produce Chloramines, again potential carcinogens.

Finally, a new process for washing vegetables is that used by the Unipolar water disinfection system. This system uses a current of electricity to split the salts present in the water to form sanitiser molecules that then act to sanitise leafy vegetables. The process is easy to use and can be incorporated easily into existing washing systems as they are found. A full commercial scale trial conducted on a grower property has shown that this system has a number of advantages over conventional sanitisers, including Chlorine. Firstly, it is automatic and continuously produces a set amount of sanitisers on a recycling system. Secondly, it records the level of sanitiser used in a continuous manner, thereby satisfying the QA requirements from a HACCP point of view. Thirdly, the shelf life of the sanitised product appears to be higher than the shelf life obtained when using other sanitisers. Finally, no chemicals need to be added to the process to achieve sanitation with the Unipolar system even though it could work better in the presence of low levels of salt.

9. Technology Transfer

A range of methods were used for technology transfer as part of this project, the most important one was direct discussions with a number of growers that have experienced issues related to washing their produce. Furthermore, some of the results from this study have been presented at a Victorian Vegetable Grower Association meeting. The results have also been presented to the Freshcare technical steering committee. It is hoped that the information gathered as part of this project will be presented as a Vege-Note in the vegetable industry. The results of this project will be used to further strengthen on-farm QA systems, including Freshcare and Salad GAP.

10. Project Evaluation

The information supplied to growers has led to a better understanding of what is available to them for washing. A lot of interest has been generated by growers with regards to the new technology trialled as part of this project, namely the Unipolar water disinfection system. Many growers have also expressed concerns that there are no plans in place should Chlorine based sanitisers be banned in the industry. This project has alleviated some of their concerns as they now know that there are alternatives available.

11. Conclusions

The washing of leafy vegetables is an important step to keep leafy vegetables safe for consumers. This step is normally part of a wider food safety plan that growers must maintain on their farms and hence is part of the QA system audited by third parties. Therefore, any sanitiser used for washing leafy vegetables must be capable of being monitored.

The use of Chlorine either as Calcium Hypochlorite or Sodium Hypochlorite is still the most effective way to sanitise leafy vegetables in Australia. There are however other sanitisers available that have similar efficacies as Chlorine, including Nylate and some of the Peroxyacetic acid sanitisers. These are however slightly more expensive to use but all of these can be monitored in solution either by hand (with the use of test strips) or some automated process including OR probes or PAA probes.

There are organic based sanitisers available for washing leafy vegetables in the organic industry, however these are not as effective as the non organic sanitisers and are difficult to monitor for the purposes of record keeping in QA food safety plans.

There is a newly available technology that uses oxidized water to sanitise vegetables that has the prospect of fulfilling the requirements for washing vegetables without the addition of harsh chemicals and is capable of exceeding a 2 log reduction in total plate counts. The use of gas based sanitisers in the vegetable industry is not recommended for a number of reasons, the most important one being OH&S.

The results of this study show that:

In terms of efficacy:

Oxidizing water>Chlorine/Nylate>Peroxyacetic acid sanitisers>Organic acid sanitisers>Acetic acid

In terms of running costs

Oxidising water<Chlorine< Acetic acid<Nylate<Peroxyacetic acid sanitisers<Organic acid

In terms of set up costs, Oxidising water is still an unknown.

12. Key Issues

• Chlorine based sanitisers are still the most cost effective and most suitable sanitisers that the vegetable industry can use to wash leafy vegetables.

• There are other sanitisers available that can be used to wash vegetables in case Chlorine is banned (as is the case in some overseas countries), which includes the Peroxyacetic acid sanitisers and Nylate.

• There is a very efficient sanitation process available that needs further evaluation which uses Oxidised water produced by passing a current through recirculated water from the wash baths. This system can be fully automated and can record every step of the washing process, thus fulfilling many of the requirements of an on-farm QA plan.

13. Recommendations for Future Work

The following recommendations are for future work:

- Develop a contingency plan in case Chlorine based sanitisers are banned in Australia.
- Develop training material for the sanitation step to be used by growers who appear confused at the best way to sanitise vegetables at the moment. Include a range of sanitisers in the training material.
- Further develop the oxidized water system for use on leafy vegetable farms. The system should be developed so that it is automatic and records every aspect of the sanitation step.
- Investigate how the microbial load of sanitized produce differs at the end of shelf life depending on the type of sanitiser used.
- Some washing technologies were not able to be comprehensively tested as a suitable system to deliver the active component in solution was unavailable for this round of testing. This included both Ozone and Chlorine dioxide. If a suitable system ever becomes available in Australia then these two technologies should also be tested and compared to other sanitisers.

14. References

Allende, A., Selma, M.V., López-Gálvez, F., Villaescusa, R., Gil, M.I., 2008. Role of commercial sanitizers and washing systems on epiphytic microorganisms and sensory quality of fresh-cut escarole and lettuce. Postharvest Biology and Technology 49, 155–163.

Behrsing, J., Winkler, S., Franz, P., and Premier, R. 2000. Efficacy of chlorine for inactivation of *Escherichia coli*. Postharvest Biology and Technology, 19, 187–192.

Beuchat, L. R. and J-H. Ryu. 1997. Produce Handling and Processing Practices. *Emerg. Infect. Dis*. 3:459-465.

Brackett, R.E., 1987. Antimicrobial effect of chlorine on Listeria monocytogenes. *J. Food Prot.* 50, 999–1003, 1008.

Department of Health Victoria guidelines (2010)
http://www.health.vic.gov.au/environment/water/recycle.htm#guidelines

López-Gálvez, F., Allende, A., Selma, M.V., Gil, M.I., 2009. Prevention of Escherichia coli cross-contamination by different commercial sanitizers during washing of fresh-cut lettuce. International Journal of Food Microbiology

López-Gálvez F., María I. Gil, Pilar Truchado, María V. Selma, Ana Allende 2010 Cross-contamination of fresh-cut lettuce after a short-term exposure during pre-washing cannot be controlled after subsequent washing with chlorine dioxide or sodium hypochlorite *Food Microbiology*, *Volume 27, Issue 2*, *April 2010*, *Pages 199-204*

Fabrizio, K.A. and C.N. Cutter. 2003. Stability of electrolyzed oxidizing water and its efficacy against cell suspensions of *Salmonella Typhimurium* and *Listeria monocytogenes*. *J. Food Prot*. 66:1379-1384.

Guzel-Seydim, Z.B., A.K. Greene, and A.C. Seydim. 2003. Use of ozone in the food industry. *Swiss Society of Food Sci and Technol*. 37:453-460.

S. J. Hooper, M. A. O. Lewis, M. J. Wilson and D. W. Williams 2011 Antimicrobial activity of Citrox® bioflavonoid preparations against oral microorganisms *British Dental Journal*, 210, E22 (2011)

Khadre, M.A., A.E. Yousef, and J.–G. Kim. 2001. Microbiological aspects of ozone applications in food: a review. J. Food Science. 66:1242–1252.

Koseki, S. K. Yoshida, Y Kamitani, S. Isobe, and K. Itoh. 2004. Effect of mild heat pre-treatment with alkaline electrolyzed water on the efficacy of acidic electrolyzed water against Escherichia coli O157:H7 and Salmonella on lettuce. Food Microbiol. 21:559-566

Koseki, S., K. Yoshida, S. Isobe, and K, Itoh. 2001. Decontamination of lettuce using electrolyzed water. *J. Food Prot*. 64:652-658.

Koseki, S. and K. Itoh. 2001. Prediction of microbial growth in fresh-cut vegetables treated with acidic electrolyzed water during storage under various temperature conditions. *J. Food Prot*. 64:1935-1942.

Kim, C., Y.C. Hung, and R.E. Brackett. 2000. Roles of oxidation-reduction potential in electrolyzed oxidizing and chemically modified water for the inactivation of food-related pathogens. *J. Food Prot*. 63:19–24.

Kim, C., Y.C. Hung, and R.E. Brackett. 2000. Efficacy of electrolyzed oxidizing (EO) and chemically modified water on different types of foodborne pathogens. *Int. J. Food Microbiol*. 61:199-207

Izumi, H. 1999. Electrolyzed water as a disinfectant for fresh-cut vegetables. *J. Food Sci*. 64:536-539

Sapers, G.M. 2001. Efficacy of washing and sanitizing methods for disinfection of fresh fruits and vegetable products. *Food Technol. Biotechnol*. 39:305- 311.

Stevenson, S.M., S.R. Cook, S.J. Bach, and T.A. McAllister. 2004. Effects of water source, dilution, storage, and bacterial and fecal loads on the efficacy of electrolyzed oxidizing water for control of *Escherichia coli* O157:H7. *J. Food Prot*. 67:1377-1383.

Shtannikov, E., V. Morozovla, and M. Morozovla. 1977. Use of electrolysed solutions of table salt for water decontamination. Gig. Sanit. 8:15-19.

Suslow, T.V. 1997. Postharvest chlorination: basic properties and key points for effective disinfection. University of California, Division of Agriculture and Natural Resources. Publication 8003

Swem, B.L. H. Yang, and Y. Li. 2002. Effects of electrochemically activated water dipping treatment on bacterial contamination of fresh-cut vegetables. In Yang, H., B.L. Swem, and Y. Li. 2003. The effect of pH on inactivation of bacteria on fresh-cut lettuce by dipping treatment with electrochemically activated water. *J. Food Sci*. 68:1013-1017.

Nakagawara, S., T. Goto, M. Nara, Y. Ozawa, K. Hotta, and Y. Arata. 1998. Spectroscopic characterization and the pH dependence of bactericidal activity of the aqueous chlorine solution. *Analytical Sciences*. 14:691-698.

Nacimento, M.S., N. Silva, M.P.L.M. Catanozi, and K.C. Silva. 2003. Effects of different disinfection treatments on the natural microbiota of lettuce. *J. Food Prot*. 66:1697-1700.

Park, C.M., and L.R. Beuchat. 1999. Evaluation of sanitizers for killing *E. coli* O157:H7, *Salmonella*, and naturally occurring microorganisms on cantaloupes, honeydew melons, and asparagus. *Dairy Food Environ. Sanit*. 19:842-847.

Park, C.-M., Y.-C. Hung, M.P. Doyle, and C. Kim. 2001. Pathogen reduction and quality of lettuce treated with electrolyzed oxidizing and acidified chlorinated water. *J. Food Sci*. 66:1368-1372.

Park, C.M., Y.C. Hung, R.E. Brackett. 2002. Antimicrobial effect of electrolyzed water for inactivating *Campylobacter jejuni* during poultry washing. *Int. J. Food Microbiol*. 72:77-83.

Venkitanarayanan, K.S., G.O. Ezeike, Y.C. Hung, and M.P. Doyle. 1999. Efficacy of electrolyzing water for inactivating *Escherichia coli* O157:H7, *Salmonella enteritidis*, and *Listeria monocytogenes*. *Appl. Environ, Microbiol*. 65:4276-4279.

Upadhyaya H., M. H. Khan, S. K. Panda 2007. Hydrogen Peroxide Induces Oxidative Stress in Detached leaves of Oryza Sativa. *Gen. Appl. Plant Physiology*, 2007, 33 (1-2), 83-95 83

Yang, H., B.L. Swem, and Y. Li. 2003. The effect of pH on inactivation of bacteria on fresh-cut lettuce by dipping treatment with electrochemically activated water. *J. Food Sci*. 68:1013-1017.

Zhang, S. and J.M. Faber. 1996. The effects of various disinfectants against *Listeria monocytogenes* on fresh-cut vegetables. *Food Microbiol*. 13:311- 321.

Appendix 1:

Table showing the criteria for evaluating visual quality of spinach and baby leaf used in this study.

Rating	***1***	***2***	***3***	***4***	***5***
Overall visual quality *(saleability and consumer appeal)*	very poor *(not useable)*	poor *(serious deterioration, unsaleable and limit of useability)*	fair *(deterioration evident, limit of saleability)*	good *(minor symptoms of deterioration, doesn't affect useability or eating quality)*	very good *(none or very slight symptoms of deterioration)*
Wetness *(moisture within product)*	severe *(>20% of leaves totally covered)*	moderate *(10-20% of leaves totally covered)*	slight *(a few drops per leaf, <20% of leaves)*	dry	none
Discolouration *(browning of cut surfaces)*	severe *(objectionable discolouration, limit of saleability)*	moderate *(obvious discolouration on most cut surfaces)*	slight *(discolouration evident on a few cut surfaces)*	trace *(just visible, not objectionable)*	none
Wilting *(individual leaves)*	severe *(>20% of leaves totally wilted)*	moderate *(10-20% of leaves totally wilted)*	slight *(minor signs of wilting – limit of saleability)*	dry	none
Rots *(on product)*	extreme *(not useable)*	severe *(serious deterioration, unsaleable and limit of useability*	moderate *(deterioration evident, limit of saleability)*	slight *(minor symptoms of deterioration, doesn't affect useability or eating quality*	None

Appendix 2:

Photographs of Unipolar Disinfection system developed to wash leafy vegetables

Photo a) The portable unit developed to deliver sanitised water to existing wash tanks

Photo b) Photo of hoses between the unit and the wash tank (the ones showing opaque water due to the formation of sanitiser chemicals are leading to the wash tank, the ones showing clear water are the hoses returning the water to the unit from the wash tank).

Photo c) Photograph of control panel including automatic reading devices that record the levels of sanitisers produced during the sanitation step.

Photo a)

Photo b)

Photo c)

Appendix 3:

Copies of newspaper clippings related to the hazards of gaseous sanitisers.

Ten hurt in toxic leak

John Watts

A HAZARDOUS chemical leak at a Clyde celery farm last Wednesday forced 10 workers to be taken to hospital.

Chlorine dioxide was released into the air at J. and J.M. Schreurs and Sons on Twyford Rd after an automated chlorinator malfunctioned.

Clyde CFA captain Ken Blencowe said that as part of a water-purification process, chlorine concentrate was fed from a pipe into a water container, but the water level was too low and the concentrate was exposed to the air.

"Chlorine in its concentrated form can be dangerous," Mr Blencowe said.

"When it comes into contact with the air it forms a gas. We had to decontaminate people before they could go to hospital."

Metropolitan Ambulance Service spokesman Andrew Watson said the workers suffered throat irritations, sore eyes and dizziness.

With most of the employees speaking another language, attending patients was a challenge. "Most of those affected were Vietnamese so we had to use interpreters to explain the decontamination process," Mr Watson said.

Eight women and two men were sent to Casey, Dandenong and Frankston hospitals. They were released that afternoon.

Emergency services at the scene of the chlorine dioxide leak in Clyde. Picture: DALE MANN. N51CS213

Market mayhem

TEN people were rushed to hospital last Wednesday after a chemical spill at a market garden in Clyde.

A Metropolitan Ambulance Service spokeswoman said the victims were overcome by fumes from a spill of a low-grade chemical, believed to be chlorine dioxide, at the Twyford Road property about 9.30am.

She said the chemical, a choking agent, caused throat and eye irritation.

The patients, eight women in their early to mid-20s and two men in their late-30s, had to be decontaminated by firefighters on site before being taken to hospital.

Interpreters were required to assist emergency crews with the patients, who were mainly of Vietnamese descent, the spokeswoman said.

Seven people were taken to Casey Hospital at Berwick, two to Dandenong Hospital and one to Frankston Hospital with minor injuries.

All were in a stable condition and would be kept in hospital for observation, she said

Members from the Metropolitan Ambulance Service, the Country Fire Authority and local police attended.