

1. INTRODUCTION

1.1. Background

Tomato is one of the most important horticultural commodity and most people use it as complementary seasoning. Tomato also can be eaten directly as fresh fruit or processed into tomato sauce, juice, paste, and others. Tomato not only has a good taste but also rich in vitamins, minerals, and bioactive compounds which is good for human health. The market demand of tomato is increasing not only for local market but also international market. In Indonesia, the production (tons/ha) of tomatoes has increased from 2015 to 2016 by 5.80% whereas the harvest area (Ha) has increased by 0.5% (BPS, 2017). The consumption of tomatoes also increased from 1065.42 million kg (2015) to 1149.16 million kg (2016) (Susenas, 2017).

There are many varieties of tomato like cherry tomato, plum tomato, round tomato, beefsteak tomato, and others. Tomato is widely consumed as raw fruit in making of salad, sandwich, burger, tomato juice, and others. Consumers of raw fruits and vegetables should be aware to the safety because the contamination of pathogenic microorganisms during transportation and the presence of pesticides or chemical fertilizer residue which can be harmful to human health. Consumers' demand and competition with globalized market imposed the needs for high standard foods. The quality of fruits and vegetables can be characterized by sensory (appearance, flavour, and texture), nutritional value, and safety attributes. Out of these attributes, appearance is the most important in determining the commercial value of the product since it is a critical factor driving initial purchase (Deliza & MacFie, 1996).

Cherry tomato is perishable food which has short shelf life in room temperature storage. On the other hand, cherry tomato has high economical value so that the cause of damage should be reduce and need improvement in post harvest handling and during storage. Accordingly, it is necessary to develop technology that can preserve cherry tomato with minimal process but give the maximum shelf life. Cherry tomato which is washed with running tap water only removes dirt like dust and soil but can not eliminate microorganisms (Brackett, 1992 in Koseki *et al.*, 2004). Washing cherry tomato using disinfectant can increase the shelf life. Disinfectant can be used to kill microorganisms

which is cause the greatest reduction of cherry tomato quality. The most common disinfectant for fresh produce is chlorine. The concentration of chlorine to wash fresh produce is 100-200 ppm (Willet *et al.*, 1989 in Okull & Laborde, 2004). But, the use of chlorine can be harmful for human health due to the residue levels. Alternatives disinfectants are therefore needed.

Study on the use of electrolyzed water (EW) as disinfectant to wash cherry tomato is rarely done, especially in Indonesia. Electrolyzed water is the product of the electrolysis of water in an electrolysis chamber, which has free chlorine as disinfectant. There are two types of EW, based on its pH i.e. acid and base (Rahman *et al.*, 2016). This research will be used both types of EW. Acidic Electrolyzed Water (AEW) is produced by anode side with $\text{pH} \leq 2.7$ and acting as strong bactericidal and fungicidal for fruits and vegetables. Base Electrolyzed Water (BEW) is generated from cathode side which produces hydrogen with $\text{pH} > 11$ and used as a cleanser and degreaser (Hricova *et al.*, 2008).

1.2. Literature Review

1.2.1. Cherry Tomato

Tomato is one of the most widely cultivated crops throughout the world including in Indonesia. Tomato not only has a good taste but also rich in vitamins, minerals, and bioactive compounds which is good for human health. The market demand for tomato is ever increasing. In Indonesia, from 2015 to 2016, the production of tomatoes increased by 5.8% and 0.5% in terms of the harvested area (Ha) and the production (tons/ha), respectively (BPS, 2017).

Tomato is included in *Solanaceae* family. Tomato was originated from Equador and Peru and then spread all over America and later spread throughout the world. In general, tomato is divided into 4 categories i.e. round tomato, cherry tomato, plum and baby plum tomato, beefsteak tomato. The main features that distinguish the four categories are the size and the shape of fruit. Cherry tomato is generally red and very sweet,

smaller than classic tomato, with weight varying between 10-20 g and diameter 1.6 – 2.5 cm (Pinheiro *et al.*, 2013).



Figure 1. Cherry Tomatoes
(Source: Private document)

According to Peralta & Spooner (2000), the classification of cherry tomato:

Kingdom	: <i>Plantae</i>
Division	: <i>Spermatophyta</i>
Class	: <i>Magnoliopsida</i>
Ordo	: <i>Solanales</i>
Family	: <i>Solanaceae</i>
Genus	: <i>Solanum L</i>
Species	: <i>Solanum lycopersicum L</i>
Variety	: <i>Solanum lycopersicum L. var cerasiforme</i>

The chemical content of tomato is influenced by several factors such as cultivars, maturity stage, temperature, climate, light, fertilizer, and irrigation. Tomato is very nutritious which is good for human health like folate, potassium, vitamin C, vitamin A, carotenoids (lycopene and β -carotene), and flavonoids. The red colour of tomato comes from the lycopene while the taste of tomato is attributed to its organic acids and sugars (Pinheiro *et al.*, 2013). The nutritional composition of red tomato fruit is shown in Table 1. The quality of tomato is also determined by colour, texture, and flavour. According to Commission Europe Union (2011), there are several classes of tomato for sale. The classification can be seen on Table 2.

Table 1. Nutritional Composition of Red Tomato Fruit

Nutrient	Value per 100 g
Water	94.52 %
Protein	88 g
Total lipid (fat)	0.20 g
Ash	0.50 g
Carbohydrate, by difference	3.89 g
Fiber, total dietary	1.2 g
Sugar, total	2.63 g
Minerals	
- Calcium	10 mg
- Iron	0.27 mg
- Magnesium	11 mg
- Phosphorus	24 mg
- Potassium	237 mg
- Sodium	5 mg
- Zinc	0.17 mg
- Copper	0.059 mg
- Manganese	0.114 mg
Vitamin	
- Vitamin C	0.860 mg
- Vitamin B	0.080 mg
- Vitamin A	42 mg
Carotene, beta	449 mcg
Lycopene	2573 mcg

Source: USDA (2010)

Table 2. Tomato Classifications for Sale

Classification	Extra class	Class I	Class II
Quality	Excellent quality	Good quality	The shape of tomato may be irregular
Hardness	Flesh fruits must be firm and have all the characteristics typical of the variety	Flesh fruits with reasonably firm	Flesh fruits with reasonably firm
Healed and Unhealed Crack	They must have no defects. No “green backs” are allowed. Fruits must be clean, free of all foreign substances	They may have slight defects and shallow scars or healed cracks* not more than 1.0 cm in length	Fruits must not have unhealed cracks. However, healed cracks* not more than 3 cm in length are allowed

Source: Commission implementing regulation (EU) N° 543 (2011)

*The first few cracks can be healed by itself (left scars) and the tomato ripen normally

Cherry tomato has cuticle as outer membrane that covers aerial parts of the plant. Cuticle is an efficient barrier to water loss, attenuates ultraviolet light absorption and protects from biotic and abiotic environmental stresses (Dominguez *et al.*, 2012). The composition of cuticle includes cutin, waxes, and polysaccharides. Cutin is a framework of cross-linked fatty acid, with waxes deposited on the surface and inside cutin matrix. The inner side of the cuticle contains various polysaccharides derived from the epidermal cell wall, which links the cuticle to the cell wall (Jeffree, 2006).

1.2.2. Deterioration of Tomato Quality

Cherry tomato that has been harvested must fulfill the acceptable quality by consumers, which it has firm and smooth texture, normal colour, and uniform varieties. Deterioration of fresh cherry tomato is influenced by several factors such as physiological, mechanical, microbiological, environment, and conditions. Cherry tomato is perishable because it has high water content ($\pm 94\%$) and still undergoes respiration and transpiration after harvest (USDA, 2005). According to Winarno (2004), respiration is oxidative breakdown of complex compounds such as starch, organic acids, etc into smaller compounds like CO_2 and H_2O . Transpiration is an irreversible loss of water from tissues due to evaporation. It will result in shrinkage, weight loss, change in texture (softening) and appearance (Pinheiro *et al.*, 2013).

Tomato quality deterioration can be caused by many factors during harvesting, post harvest handling, distribution, and storage. Fully ripe tomato is very susceptible to the mechanical damage which will shorten the shelf life. The harvested produce contains substantial amount of heat, called as field heat. Pre-cooling stage aims to eliminate excessive heat, thereby reducing microorganism's activity, to decrease respiration and metabolism rate, as well as decrease ethylene production. Pre-cooling stage can be done by using cold water mixed with disinfectant so it can extend the shelf life. The use of disinfectant either for cooling or washing is expected to reduce the amount of pathogenic microorganisms in produce, such as tomatoes.

Tomato is rich in nutrition and contains much water so it is very susceptible to the microbial damage. Microorganisms can attack fresh produce and spread rapidly because

of the abundance nutrients and moisture that support their growth. Tomato is more susceptible to fungi than bacteria (Barth *et al.*, 2009). Fungi that often attack tomato fruit are *Aspergillus sp*, *Fusarium sp*, *Rhizopus stolonifer*, *Penicillium notatum*, *Mucor sp*, *Botrytis cineria*, *Rhodotorula sp*. Besides fungi, tomato can be attacked by bacteria such as *Lactobacillus fermenti*, *Pseudomonas stutzeri*, *Listeria monocytogenes*, *Leuconostoc sp*, *Rothia sp* (Mbajiuka *et al.*, 2014).

Colour is one of the most important external characteristics in determining consumer acceptance. Cherry tomato generally have red colour that not only shows the maturity stage but also the desired level of flavour (Pinheiro *et al.*, 2013). Carotenoids such as β -carotene and lycopene contribute about 7% and 87% red colour of tomato, respectively. Lycopene (red colour) will accumulate gradually on ripening stage therefore the colour is become darker as increasing storage period. The lightness of tomato will significantly change during storage (Khairi *et al.* 2015). Higher temperature storage will accelerate the colour change in tomato (Tadesse *et al.*, 2015).

Texture, especially hardness is one of the most important factors to determine cherry tomato quality. Texture may be the final consideration of consumers before purchasing tomato, they often use finger to test tomato texture at the time of selection (Batu, 1998.) Changes of cherry tomato texture during storage can be influenced by flesh hardness and skin strength. The fruit texture will become softer during storage and distribution. Ripening process can be a serious problem that can increase the rate of decay (Pinheiro *et al.*, 2013). Tomato tissues softening can be caused by weight loss, loss of turgor, and or due to enzymatic activity such as *pectin methylesterase* (PME, E.C.3.2.1.11) and *polygalacturonase* (PG, E.C. 3.2.1.15). The texture of fruit become softer when the weight loss is high (Alia-Tejacal *et al.*, 2007).

According to Ramaswamy (2015), there are two causes of post harvest loss i.e. primary and secondary causes. Primary causes of losses include biological, microbiological, physical, mechanical, chemical, biochemical, and physiological. Biological and microbiological loss can be caused by insect, pest, animals, and microorganisms (fungi and bacteria). Physical loss happened because improper environmental and storage

conditions. Chemical and biochemical loss can be caused by undesirable reactions between chemical compounds in food such as browning, enzymatic change, etc. Mechanical loss is damage caused by abrasion, crushing, bruising, puncturing, and so forth. Physiological loss happens because senescence, sprouting, and other respiratory and transpiratory changes. Secondary causes usually occur because improper harvesting and handling, inadequate storage facilities, inadequate transportation and marketing system, etc. According to USDA (2005), the characteristics of damaged tomato are abnormal colour, abnormally soft texture with unusual watery appearance, occurrence of spots, bruise, and or crack on the surface of fruit.

Fruit cracking can decrease the quality of fruit and reduce its marketability especially because cracks lead to poor appearance, reduced shelf life, and susceptible to fungal infection (Peet, 1992). Cracks occur when the internal pressure exceeds the breaking stress of the epidermis, especially the cuticle. In cherry tomato, cracks only appear during ripening. Fruit cracking and biochemical properties of cuticle are affected by environmental conditions, mainly high relative humidity (RH) and high temperature. Both conditions can decrease the elasticity of the cuticle so it will increase the cuticle deformability and the cuticle become less resistant to stress (Dominguez *et al.*, 2012).

Since fruits and vegetables are still alive after harvest, the physiological processes continue such as respiration and transpiration. Respiration results in produce deterioration and nutritional value loss. Changes in texture, flavour, and weight loss occur because of transpiration process. Water loss means weight loss and affects the appearance and texture. These process can not be stopped but the can be reduced significantly by careful management of temperature and relative humidity during storage and transportation (Ahmad and Siddiqui, 2016). According to Yahia & Brecht (2012), tomato should be stored in RH of 85-95% so the transpiration process can be minimized. Leonardi *et al.* (2000) stated that higher temperature of storage will increase the vapour pressure difference between fruit and surrounding air. If the pressure difference is high, it would accelerate the movement of water vapour from the fruit into surrounding air.

Packaging is an essential component in the food system and often used as a tool to extend shelf life by preventing or reducing water loss, especially in fresh produce. The functions of packaging are maintaining the quality, reducing postharvest losses, and protecting food from physical, chemical, and microbiological damage therefore the shelf life of the product can be extended (Opara and Mditshwa, 2013). For fruit and vegetables, packaging can not prevent from spoilage but incorrect packaging can accelerate spoilage. The choice of packaging type depends on the shape and the perishability of the product. Soft fruits like berries and cherries are highly perishable and easily to be spoiled by anaerobic microorganisms. They can bruise and crush easily which lead to spoilage. Thus, they are packaged in semi-rigid container and sometimes use ventilation holes to avoid fogging. The packaging material is usually made from PET which is suitable for varied uses. It has very high mechanical strength, light weight, and transparent. PET is resistant to chemical compounds, stable over a wide range of temperature (-60°C to 220°C), and have adequate gas barrier properties (Shin and Selke, 2014).

1.2.3. Total Plate Count (TPC) and Direct Plating

The principle of TPC method is growth the living microorganisms in agar media so that microorganisms can multiply and forms colonies that could be seen directly without using microscope. Before the microorganism is grown in media, the sample must be diluted. The dilution is used in order to reduce the number of microbes in the sample (Sutton, 2011). Calculation of TPC is expressed as the number of microorganisms colonies multiplied by dilution factor. TPC method was performed on petri dish that had colonies between 30 and 300. Microbial colonies can be counted after incubation process which generally takes 24 hours or more (Aryal, 2016).

Nutrient Agar (NA) media was used to grow bacteria and Potato Dextrose Agar (PDA) media was used to grow fungi. The composition of NA media includes extract beef, peptone, and agar. Agar is a polysaccharide extract from marine algae and act as a solidifying agent used in microbiological media. Agar melts at 84°C and solidifies at 38°C. Peptones are hydrolyzed protein formed by enzymatic or acidic digestion and as nitrogen source. Extract beef is used as nutrients source for the cultivation

microorganisms and contains high concentration of water soluble protein and glycogen. In contrast to NA, PDA media uses plant extract (potato) to enrich the nutrition of media. Potato extract contains high concentration of carbohydrate. Besides potato extract, PDA contains dextrose and agar. Dextrose is sugar clusters (monosaccharide or polysaccharide) that acts as nutritional enhancer (Atlas, 2010).

Cherry tomatoes are harvested without removing the stem and calyx. Calyx is the entrance of germ to attack the tomato through the calyx's stomata so the germs can passage easily into the fruit. Fungi usually appear on the calyx of tomato. The highest bacterial and fungal counts were detected on tomato's calyx. Therefore, calyx is the main source of potential spoilage microorganisms of tomato (Khalegi *et al.*, 2013). The main defect of cherry tomato is mouldy stem and or decayed stem. Mould that affecting the tips of stem or edges of the calyx are considered objectionable when detracting the appearance (USDA, 2005).

1.2.4. Chlorination

Chlorine has been used widely as disinfectant in production, harvesting, post harvest handling, and marketing display of fresh fruits and vegetables. Chlorine contributes to the inactivation of pathogenic bacteria, viruses, fungi, and spores of microorganisms in fresh produce. However, chlorine can be harmful for human health due to the residue levels. If the organic materials were not incompletely oxidized, the leftover chlorine will produce undesirable by products that have carcinogenic potential at high dose, such as chloroform (CHCl_3) or other trihalomethanes. Chlorine also reacts with organic nitrogen-based materials to produce mildly toxic chloramines at high pH (Richardson *et al.*, 1996 in Suslow, 2000).

There are three forms of chlorines that legally used in agriculture i.e. chlorine gas (Cl_2), calcium hypochlorite (CaCl_2O_2), and sodium hypochlorite (NaOCl). The most common form of chlorine used for disinfection of produce is calcium hypochlorite (CaCl_2O_2). Besides used as disinfectant, calcium hypochlorite (CaCl_2O_2) can extend the shelf life and improve disease resistance of fruits and vegetables by adding calcium to the cell wall. Calcium in chlorine can also enhance the cell wall integrity, protect from

degrading enzymes, and increase the cohesion of tomato's cell wall (Pinheiro *et al.*, 2013). The common concentration of chlorine for washing fresh produce is 100-200 ppm which can kill bacteria and fungi spores (Willet *et al.*, 1989 in Okull & Laborde, 2004). The antimicrobial form, HOCl, is higher at pH 6.5 to 7.0 (exists as 80-95%). At higher pH (>8.0), the level drops to less than 20%. This shows that the effectiveness of disinfectant is much less at high pH (Suslow, 2000).

1.2.5. Electrolyzed water (EW)

Electrolyzed water is a product of the electrolysis of water in an electrolysis chamber, which has free chlorine as disinfectant. Tap water contains chloride therefore it is possible to obtain reactive free chlorine in tap water by electrolysis. The EW generator usually electrolyzed at low voltage value of 10-20 V. The electrolysis apparatus consist of two electrodes, known as anode and cathode (Koseki *et al.*, 2003). There are several factors that affect the effectiveness of EW for disinfection. The main factors are the concentration of free chlorine, pH, and Oxygen Reduction Potential (ORP). The other factors are current, water flow rate, salt concentration, storage condition, and so on (Rahman *et al.*, 2016).

EW has numerous advantages such as the possibility for on-site production, environmentally friendly because no chemical added, more effective than chlorine in killing microorganisms, low cost and safe, and can maintain the sensory quality of foodstuffs (Rahman *et al.*, 2016). However, there are some disadvantages of EW that need to be considered including the high initial cost of equipment, the tendency for EW to lose its antimicrobial potential quickly because of improper storage, the presence of organic materials, or the decreasing free chlorine concentration over time. Furthermore, EW with very low pH (AEW) might be corrosive and irritating for hands (Rahman *et al.*, 2016). The antimicrobial efficacy of EW is also highly influenced by storage conditions. Under open condition, the loss of chlorine concentration will be greater because of evaporation. EW kept under closed condition will loses chlorine by self-decomposition. The loss of chlorine under closed condition is lower than that of evaporation under open condition (Len *et al.*, 2002).

The active chlorine species, known as Cl_2 , HOCl , and OCl^- play important role in inactivation of microbial cells. The other oxidants such as ozone and hydrogen peroxide are produced during electrolysis also have antimicrobial activity in EW. The first step of disinfection is performed by OCl^- . Ionized OCl^- will rupture the microbial cell wall but unable to penetrate the cell membrane because of the existence of the lipid bilayer. Ionized OCl^- only attack on the outer membrane and has shown poor germicidal activity. On the other hand, HOCl can penetrate into the microbial cell membrane and then pass the lipid bilayers of plasma membrane by passive diffusion. HOCl can attack on the outer membrane and also within the cell. As a result, HOCl and OCl^- have germicidal action by destroying the membrane and DNA of microorganisms, inactivate the enzyme activity that essential for microbial growth, and might deteriorate membrane transport capacity (Rahman *et al.*, 2016).

Based on its pH, there are two types of EW, i.e. acid and base (Hricova *et al.*, 2008). Base Electrolyzed Water (BEW) is generated from cathode side which produces hydrogen with $\text{pH} > 11$ and ORP value between -800 mV and -900 mV which used as cleanser and degreaser. BEW that has pH from 8 to 10 called Slightly Alkaline EW. Acidic Electrolyzed Water (AEW) is produced by anode side with $\text{pH} \leq 2.7$, ORP value > 1100 mV, and chlorine 10-90 ppm. AEW produces chlorine and then reacts with water to form HOCl and HCl . AEW plays a role as strong bactericidal. AEW also contributes to the inactivation of bacteria and fungi in fresh fruits and vegetables (Rahman *et al.*, 2016). AEW is divided into two categories i.e. Strong Acid EW and Slightly Acid EW. Strong AEW has pH from 2 to 3 and high ORP (1100 mV) and Slightly AEW has pH about 5-6.5 with ORP value 850 mV.

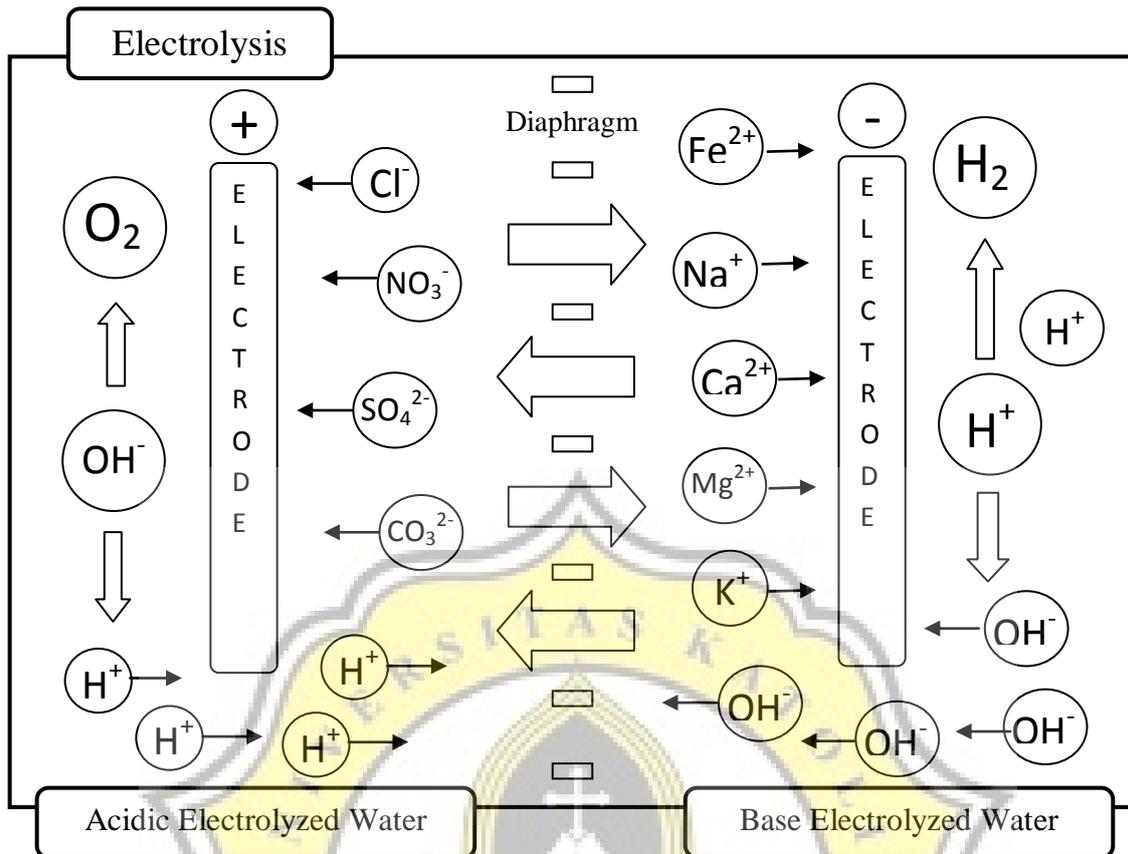


Figure 2. Production of AEW and BEW in Electrolysis Cell

Production of AEW and BEW in electrolysis cell, consisting of anode and cathode connected through an external power supply. ORP seems to be main contributors to the antimicrobial activity of EW. ORP is defined as an indicator showing the ability of oxidation or reduction of a solution. The higher ORP value the higher oxidation capability. The ORP value is also affected by pH where the lower pH leads to the higher ORP value. The higher ORP value is usually followed by higher chlorine residues that have antimicrobial activity (Park *et al.*, 2004). However, ORP is not the primary factor affecting the disinfection process (Koseki *et al.*, 2004).

DO (Dissolved Oxygen) is an indicator of water-soluble oxygen content in water. The DO value is one of parameters used to determine water quality. The dissolved oxygen in water is necessary for aquatic organisms to grow and survive. There are several factors that affect DO value such as atmospheric pressure, temperature, water flow rate, and

pollution (Lewis, 2006). AEW has high DO value and BEW has low DO value (Huang *et al.*, 2008 and Rubik, 2011).

E. coli is the most resistant to BEW, quite resistant to slightly AEW, and not resistant to AEW (Rahman *et al.*, 2016). The resistance level of bacteria is affected by growth phase where bacteria spores highly resistant than vegetative cells (Rahman *et al.*, 2012). Moreover, dipping time can influence the efficacy of EW. According to Koseki *et al.* (2004), dipping for 10 minutes in strong AEW can reduce the number of microorganisms effectively i.e. 1-2 logs of aerobic mesophilic bacteria, coliforms, and fungi. Additional treatment like agitation can increase the efficacy of EW especially AEW to inactivate microorganisms. Chlorine residues in EW can be spread throughout the fruit surfaces and release microorganism cells that attached on the surface area. The microorganism cells that released from the surface will become less resistant so the inactivation will be easier (Park *et al.*, 2004). The agitation treatment will reduce chlorine residues in EW up to 5 times because more chlorine residues that react with microorganism cells (Len *et al.*, 2002).

1.3. Research Objectives

The objectives of this research was to investigate the efficacy of different washing solutions (tap water, chlorinated water, AEW, and BEW) in extending the shelf life of cherry tomato and maintaining the quality of cherry tomato based on the physicochemical and microbiological characteristics.