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Ethylene as a Contaminant in the Industrial Horticultural Distribution Chain

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Abstract

Ethylene is a hormone, which participates in the maturation of horticultural products. High ethylene concentrations can cause the senescence of products, so ethylene management is important in the supply chain to extend postharvest shelf life. In this study, the ethylene concentration in 6 different locations was monitored in Manawatu, New Zealand. A distribution centre was found to have a higher environmental ethylene concentration than supermarkets, possibly due to the combustion engine exhausts. A room with high ethylene production products, such as apples and avocado, also had the highest ethylene concentration (3670 nL L^{-1}). For the supermarkets, 80% of the time ethylene concentrations in the chiller room was below 100 nL L^{-1}. However, more ethylene producing products were stored in the chiller room of one supermarket (B), and its 80^{th} percentile ethylene concentration was 207 nL L^{-1}. Contrastingly, ethylene concentrations within a flower store were measured to be as low as the ambient environment.

Broccoli (*Brassica oleracea* var. *italica*) is a popular vegetable with short shelf life. Ethylene can induce changes in quality of broccoli, such as yellowing. After being informed of potential ethylene concentrations in the supply chain, five continuous ethylene contamination treatments (0, 50, 100, 500, 1000 nL L^{-1}) were applied to broccoli for two weeks, in order to quantify consequent quality effects. Broccoli exposed to > 500 nL L^{-1} was yellower than other treatments. The increase of chroma (C) and decrease of hue angle (h) and lightness (L) were faster than low ethylene concentrations (0 and 50 nL L^{-1}). However, there were no significant differences in colour degradation between 50 nL L^{-1} and the control. The effects to

broccoli exposed to 100 nL L^{-1} ethylene treatment was intermediate between 50 and 500 nL L^{-1}.

The broccoli was sensitive to > 100 nL L^{-1} ethylene. The 80th percentile of ethylene concentration in distribution centre was greater than 100 nL L^{-1}, with the peaked measured being more than 1000 nL L^{-1}. For supermarkets, although the ethylene concentration was below 100 nL L^{-1} in most supermarkets, the peak measurement can be higher than 100 nL L^{-1} (even more than 300 nL L^{-1}). It took around 12 days to make difference. There is potential for broccoli to be affected by the current ethylene environment in the supply chain. Therefore, in these scenarios, there is a potential justification for ethylene management to result in improve quality delivery and extension of product storage life.

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List of Abbreviations

a	Redness/Greenness
ACC	1-aminocyclopropane-1-carbox-tlic acid
ACO	ACC oxidase
ACS	1-aminocyclopropane-1-carboxylase synthase
AVG	Aminoethoxyvinylglycine
b	Yellowness/Blueness
C	Chroma
C_2H_4	Ethylene
CO_2	Carbon Dioxide
GSCM	Global supply chain model
h	Hue angle
KCl	Potassium chloride
L	Lightness from black to white
MTA	5’-deoxy-5’methylthioasenosine
1-MCP	1- methylcyclopropene
O_2	Oxygen
PMMT	Protonated montmorillonite
RH	Relative Humidity (%)
SAM	S-adenosylmethionine
TCR	Temperature-controlled room

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Chapter 1 Literature Review

1.1 The fresh produce supply chain

Global supply chain model (GSCM) was including a production, distribution and vendor network, and it avoid the damage of products and estimate the local demand by minimizing cost, weighting cumulative production and distribution time (Arntzen et al., 1996). Nowadays, innovative technologies and strong brand, which was used to support the supply chain, was the winning combination, as it can meet the market demand and the changing conditions (ElMaraghy & Mahmoudi, 2009). For the system of the supply chain, the production of horticultural products is the first step in supply chain, and it flows by grading, packaging, the storage and transport to the market and distribution centre (Heron et al., 2001; Aitken et al., 2005). Moreover, the industries provide protocols of ethylene to avoid the deterioration and above 0 ℃ was recommended as the standard to avoid the chilling injury (Aitken et al., 2005). The information technology to monitor the products and the vertically integrated structures to control the operations were applied in New Zealand supply chain (Aitken et al., 2005).

1.2 Ethylene

1.2.1 Ethylene and physiology of horticultural products

Ethylene (C_2H_4) is a simple hydrocarbon that occurs as an odourless and invisible gas (Martinez-Romero et al., 2007). It can be measured from low concentration in parts-per-million (ppm, $\mu L\ L^{-1}$) to parts-per-billion (ppb, $nL\ L^{-1}$) (Keller et al., 2013). Wheeler et al. (2004) estimated that the effects of ethylene on plants have studied nearly 100 years. Ethylene is a ripening phytohormone that is involved in the development and growth of horticultural products and is highly associated with the maturation and stress of the plants (Janssen et al., 2013; Argueso et al., 2007; Zhang et al., 2017). The application of ethylene can be used to promote ripening of many fruit and senescence of vegetables. There is no doubt that ethylene can induce changes in products and has both negative and positive impacts on storage life of plant products in the market (Gwanpua et al; 2018). Ethylene can have positive effects, assisting in achieving desired flavour, colour, and texture of the horticultural products by stimulating ripening (Martinez-

Lerud et al., 2019). While ethylene application can be beneficial, on many other occasions undesirable concentrations of ethylene can lead to detrimental effects. Martinez-Romero et al. (2009) reported that ethylene was identified as the causative agent of plant senescence and defoliation. Also, Wills et al. (2000) stated that accumulative of ethylene can cause undesirable physiological disorders, excessive softening and chilling injuries in horticultural products. Uncontrolled ethylene can cause bitterness of vegetables, chlorophyll loss, wilting of flowers and premature ripening of horticultural products (Saltveit, 1999; Keller et al., 2013; Smith, et al., 2009; Cai et al., 2019). Therefore, keeping ethylene at a desirable concentration has the potential for maximising the shelf life of postharvest horticultural products.

1.2.2 The source of ethylene

As ethylene can be produced in large quantities within an environment, such as traffic corridors, petrochemical sites and horticultural areas, there are increasingly more studies that have showed an interest or research in ethylene concerning plants (Høyer, 1995; Morgott, 2015). Ethylene can be released from both natural and anthropogenic sources (Sawada & Totsuka, 1986; Høyer, 1995; Morgott, 2015). It has been reported that 74% ethylene can be released from the nature, while 26% can be emanated from the anthropogenic source (Sawada & Totsuka, 1986; Keller et al., 2013).

Figure 1.1 The natural (terrestrial and biomass burning) and anthropogenic sources (aquatic, fuel oil, garbage, coal combustion and leakage from industries) of ethylene (Keller et al., 2013).

As mentioned early, the air pollution, such as traffic, smoke, oil, coal combustion and welding, can cause the build-up of ethylene (Morgott, 2015). The example of ethylene concentrations, which were released by different anthropogenic sources, has been shown in the table below.

Table 1.1 Example of ethylene concentration from different anthropogenic sources (Keller et al., 2013).

Source	Ethylene concentration ($nL\ L^{-1}$)
Butane fuelled forklift exhaust	1.5×10^5
Diesel motor exhaust	6×10^4
Gasoil motor exhaust	2×10^5
Cigarette smoke	1×10^5-2×10^5

1.2.3 Biosynthesis of ethylene and mode of action

The biosynthesis of ethylene (Figure 1.2) is a highly regulated process. Firstly, the amino acid methionine can release ethylene which can be converted to S-adenosylmethionine (SAM) through S-adenosylmethionine synthetase (Zhang et al., 2017). This process needs the consumption of ATP and the addition of adenine (Argueso, et al., 2007). And then 1-aminocyclopropane-1-carboxylase synthase (ACS) with the generation of 5'-deoxy-5'methylthioasenosine (MTA) can transfer SAM into 1-aminocyclopropane-1-carbox-tlic acid (ACC), which can be recycled to methionine (Martinez-Romero et al., 2007). This step is the rate-limiting step in ethylene biosynthesis. Thus, high rates of ethylene can be achieved with a small pool of methionine. (Adams & Yang, 1979; Agarwal et al., 2012). Moreover, it seems that the ethylene, CO_2 and cyanide can be converted from ACC via the function of ACC oxidase (ACO) (Sun et al., 2017).

Figure 1.2 The biosynthesis pathway of ethylene (Martinez-Romero et al., 2007).

Meanwhile, ETR1, ETR2, EIN4, ERS1, and ERS2 are ethylene receptors. They play an important role in the mode of action of ethylene (Martinez-Romero et al., 2009; Chen et al., 2018). Over the past decade, the using of Arabidopsis is a good model to identify the mode of action of ethylene (Tang et al., 2018). The overall trend of the action is that the ethylene as a hormone needs to bind a receptor through a complicated mechanism to activate the biological responses in the plants, although the signalling components are still unknown (Chen et al., 2018). Most research studies reported that ethylene receptors, including ETR1, ETR2, EIN4, ERS1 and ERS2, will be active when ethylene absent (Serek et al., 2006). Moreover, it should be known that ETR1 and ERS1 can act on CTR1 protein directly, but other receptors cannot (Burg & Burg, 2018). Additionally, CTR1 and ethylene can act as negative regulars in the pathway (Binder, 2008). Therefore, ethylene can bind and inactive the receptors and CTR1 when it appeared (Figure 2). Meanwhile, EIN2 can change the active form and produce ethylene responses in the plants as the inactivation of CTR1 (Prange & Delong, 2003; Tang et al., 2018). However, if the alleles can be changed and the receptors cannot bind ethylene.

It possibly the CTR1 can still stay active (Burg & Burg, 2018). In addition, the insufficient of ethylene receptors can cause the insufficient activation of CTR1 and ethylene response (Prange & Delong, 2003). Therefore, the application of the action of ethylene can provide an idea to minimise the damage of ethylene.

1.2.4 Ethylene control methods

Nowadays, most research studies the method to inhibit ethylene. 1-MCP (1-methylcyclopropene) can be a potent ethylene antagonist to prevent post-harvest effects of ethylene, considering it is safe, economical, easy-applying and non-toxic (Çelikel et al., 2002; Chamani et al., 2005). Moreover, 1-MCP can minimize the damage of ethylene effectively, as it can bind the ethylene receptors, such as ERT1, ERT2, EIN4, ERS1 and ERS2, with 10 times more affinity to suppress ethylene response pathway (Martinez-Romero et al., 2007). In this way, it can keep CTR1 in the inhibiting stage effectively (Prange & Delong, 2003; Tang et al., 2018). Another chemical inhibitor – AVG (aminoethoxyvinylglycine) can inhibit ethylene, as it can inhibit the formulation of ACC (Blanke, 2014; Sun et al., 2016). The using of ethylene scrubber, such as a $KMnO_4$-based innovative C_2H_4 scrubber (using a protonated montmorillonite (PMMT)) and Bi-On® R12 scrubber, also can be an effective method to control ethylene in transport, packages and fruit stores (Álvarez-Hernández et al., 2019).

1.2.5 Climacteric and non-climacteric physiology

It has been observed that the products can be classified into climacteric and non-climacteric categories (Chen et al., 2018). Most studies qualified that the patterns of ethylene and CO_2 are the criteria to distinguish climacteric and non-climacteric products (Payasi & Sanwall, 2009; Paul et al., 2012; Symons et al., 2012). Non-climacteric behaviour can be underlined via a negative ethylene feedback mechanism (ethylene did not induce its own synthesis), while ethylene can induce its own synthesis that is climacteric behaviours (Atta-Aly et al., 2000). It is clearly that there are developmental changes, such as colour, fruit softening, and textural changes, from ovary to mature of both climacteric and non-climacteric fruits, due to the accumulation of ethylene (Saladié et al., 2007; Handa et al., 2011). However, some different influences have been found during the postharvest storage via the accumulation ethylene in both climacteric plants and non-climacteric plants.

The ripening can be stimulated via ethylene during the supply chain is one main physiological effect of the accumulation of ethylene (Yang et al., 2013; Blanke, 2014). For climacteric plants, the ripening process will be accelerated via the autocatalytic ethylene synthesis of ethylene causing quality loss of horticultural products (Argueso, et al., 2007; Binder, 2008). Wills et al. (2000) found that the ripening of climacteric fruits and vegetables can be initiated by ethylene undesirably during storage and transport, although ethylene can be applied to uniformly ripen fruit commercially, such as banana. Furthermore, Janssens et al. (2013) also claim that ethylene synthesis is an irreversible ripening process in climacteric products, and it can lead to prematurely ripened products and their decay with few exceptions of climacteric fruits and vegetables.

By comparison to the climacteric products, there was a less progress that has been made in the regulatory mechanisms of ripening in non-climacteric fruits (Chervin et al., 2004; Trainotti et al., 2005). Ethylene can promote the senescence, which is often associated with loss of green skin coloration, considering the chlorophyll will degrading, such as citrus, lemons and easy peelers (Cherial et al., 2013). For non-climacteric plants, such as eggplant, pepper, and grape, ethylene may also have negative influences, which include the reduction of postharvest life, the appearance of physiological disorders, plant senescence and the evolution of pathogen (Martinez-Romero et al., 2009). Furthermore, Wills et al. (2000) suggested that ethylene can promote senescence and the development of decay microorganisms.

The different physiological effects of the accumulation of ethylene between climacteric and non-climacteric products have been shown in the table below (table 1.2).

Table 1.2 Physiological effects of accumulation of ethylene with horticultural products between different climacteric classifications (Janssen et al., 2013).

physiological parameter	climacteric fruit	non-climacteric fruit
Ethylene synthesis	enhanced	no effect
Autocatalytic	enhanced	no effect
Fruit metabolism and respiration	enhanced	no effect
Fruit ripening	enhanced	no effect
Loss of green skin colour	Negligible effect	often enhanced
Decay microorganisms	No clear effect	often enhanced
Induction of disorders or chilling injury	possible	possible

Traditionally, fresh produce can be classified as climacteric and non-climacteric due to their different respiratory patterns (Azzolini et al., 2005; Biale, 1964). Generally, climacteric products are the main source of ethylene (Janssen et al., 2013). An ability to autocatalytically produce ethylene during ripening differentiates climacteric from non-climacteric fruits. Climacteric fruits go through a ripening process which is accompanied by an increase in respiratory rate and elevated ethylene production. This autocatalytic ethylene synthesis is an irreversible ripening process (Janssen et al. 2013). The ethylene can drop dramatically during the post climacteric phase (Azzolini et al., 2005). Giovannoni (2001) hypothesised that ethylene can play a significant role in both physiological and biochemical changes during fruit ripening. Climacteric fruits can themselves be a major source of ethylene in all parts of the supply chain including distribution centres, supermarkets, storage areas and display shelf (Janssen et al., 2013).

On the other hand, non-climacteric fruit do not have an increase in both ethylene and respiration during ripening and usually undergo a decline in respiration (Pech et al., 2008). Some examples of the general climacteric and non-climacteric fruits are listed in Table 1.3 and Table 1.4, showing the ethylene production of different fruits and vegetables.

Table 1.3 Ethylene concentration and classification of selected fruits during ripening (Burg & Burg, 1962).

Climacteric fruit	nL L^{-1}	Non-climacteric fruit	nL L^{-1}
Apple	2.5×10^4-2.5×10^6	Lemon	100-200
Pear	7×10^4-8×10^4	Lime	300-2×10^3
Peach	10^3-2.1×10^4	Orange	100-300
Avocado	2.9×10^4-74×10^4	Pineapple	160-400
Mango	50-3×10^3		
Passion fruit	4.66×10^5-5.3×10^5		
Plum	200-300		

Table 1.4 Ethylene efflux and sensitivity and classification of selected fruits (+++,very large efflux:>100 µl kg-1 h-1; +, large efflux: 10-100 µl kg-1 h-1; 0, intermediate efflux: 1-10 µl kg-1 h-1; -,less efflux:0.1-1 µl kg-1 h-1; ---, minimal efflux:<0.1 µl kg-1 h-1) (Janssen et al., 2013).

commodity	Commodity classification	Ethylene efflux	Ethylene sensitivity
apple	Climacteric	++	+
avocado	Climacteric	+	+
banana	Climacteric	0	+
carrot	non-climacteric	---	+++
citrus	non-climacteric	---	0
kiwi	Climacteric	-	+++
pear	Climacteric	+	+
passion fruit	Climacteric	+++	+
tomato	Climacteric	0	+
onion	non-climacteric	---	-

Ethylene efflux of ripening climacteric horticultural products generally exceeds that of non-climacteric products. Janssen et al. (2013) hypothesised that the ethylene efflux can be relative to the internal ethylene concentration in the fruit core rather than the ethylene sensitivity. The sensitivity of horticultural products is different among the diversities of fruits and vegetables. In Janssen et al. (2013)'s experiment, they found that the ethylene production of carrot and kiwi are 0.1 µl kg^{-1} h^{-1} and 1 µl kg^{-1} h^{-1} respectively, but both of them are quite sensitive to the ethylene (shown in Table 1.5). Another example is that 300 nL L^{-1}ethylene concentration can cause the decay of kiwifruit, while 100 nL L^{-1} ethylene level can cause the unpleasant taste and isocoumarin synthesis of carrots (Fernandez-Lafuente et al., 1996).

Table 1.5 The ethylene production of different horticultural products (Smith et al., 2009).

Low (< 1.0 ml kg-1 h-1)	Moderate (1-10 ml kg-1 h-1)	High (10-100 ml kg-1 h-1)	Very high (> 100 ml kg-1 h-1)
Pineapple	Banana	Apricot	Apple
Artichoke	Mango	Nectarine	Avocado
Cauliflower	Plum	Pear	Cherimoya
Broccoli	Tomato	Peach	Passion fruit
Celery			
Lemon			
Onion			
Asparagus			
Spinach			
Beetroot			
Orange			

Many articles claim that climacteric and non- climacteric fruits have different responses to exogenous ethylene (Azzolini, 2005; McMurchie et al., 1972). It seems that the application of exogenous ethylene (especially < 100–10^3 nL L^{-1}) can advance the respiration and the ethylene production for climacteric fruit, whereas non-climacteric fruits can only be led to a transitory respiration response (Azzolini et al, 2005; Smith et al., 2009). One example is that 10 and 100 μL L^1 of exogenous ethylene can initiate the ripening of bananas, mangoes, honeydew melon, stone fruits and kiwifruit (Wills et al., 2001; Saltveit, 1999). However, the lowest level of ethylene that can advance the ripening of climacteric fruit has not been widely explored and remains unclear. One study demonstrates that an exogenous ethylene concentration of 100-500 nL L^{-1} can be the threshold level to advance ripening in honeydew melon and pear (Knee, 1985). Moreover, the softening of kiwifruit can be initiated by extremely low ethylene concentration, which is 10 nL L^{-1} of exogenous ethylene (Mitchell, 1990; Mworia et al., 2012). Meanwhile, the banana can be exposed to >100 nL L^{-1}level of ethylene for the short term, while for long term application of ethylene, ripening can be advanced when exposed to ethylene concentration of <100 nL L^{-1} (Wills et al., 2001).

Climacteric fruits, such as banana, avocados, and pears, can ripen after harvest with physio-chemical changes including changes in colour, sweetness and softening

(Smith et al., 2009). Non-climacteric fruits such as grapes and pineapples do not ripen after the harvest, but changes such as shrinkage, rotting and discolouration can occur. Furthermore, the climacteric fruits can produce ethylene, while non-climacteric cannot do that (Ludford, 2003). Whereas, non-climacteric products can gain senescence by the effect of ethylene (Wills et al., 2001). Wills et al. (1999) estimated that the ethylene, which is >5 nL L^{-1} can reduce the commercial conditions, as they find that time to ripened increased linearly as there was a deleterious linear response to log increase and the fruits can show the greater postharvest life. There is not an effective threshold level of non-climacteric fruits (Wills et al., 2001).

1.2.6 Ethylene sensitivity in different plants

1.2.6.1 Ethylene sensitivity in vegetable and fruits

Although some horticultural products do not produce ethylene during the ripening, they still can have high sensitivity to ethylene. Table 1.6 summarises some examples of ethylene sensitivity of horticultural products to ethylene concentration.

Table 1.6 Ethylene sensitivity of horticultural products (Smith et al., 2009).

Product	<10 nL L^{-1}	<100 nL L^{-1}	≥1000 nL L^{-1}
Vegetable		Broccoli Lettuce Cabbage Carrot	Cucumber
Fruit	Kiwifruit	Tomato Strawberry	Apple Pear Avocado Orange

There is a number of common ethylene sensitive fruits and vegetables that have been recorded by quantitating studies. There are some common fruits and vegetables belong to ethylene sensitive products, including apples, avocados, bananas, broccoli, cucumber, grapes, eggplant, tomatoes, onions, kiwifruit, pears, peaches, watermelon, sweet potatoes (Smith et al., 2009; Han, 2010; Bender, 2014; UCSD, 2017). At the same time, apples, avocados, bananas, kiwifruit, pears, peaches, peppers can produce ethylene too (UCSD, 2017). One thing should be taken into consideration is that the display of ethylene sensitive products should not be near to the ethylene producers (Bender, 2014; UCSD, 2017). Furthermore, ethylene producers should not be stored in bags or sealed containers (UCSD, 2017). Otherwise, it can speed up the process of ripening and advance the senescence phenomenon of these products. Ethylene insensitive horticultural products include blueberries, cherries, beans, garlic, oranges, potatoes, strawberries, tomatoes and grapefruit (Han, 2010; UCSD, 2017). The distinguishing of ethylene sensitive products and ethylene producing products can help supply chain personnel to properly utilise the display area of horticultural products. Proper handling of fresh produce in the supply chain can help prolong the storage life of fruits and vegetables.

1.2.6.2 Ethylene sensitivity in flower

Additionally, cut flowers can be classified as sensitive and insensitive too (Reid & Wu, 1992; Wouter & Doorn, 2002). The classification of some flowers is shown in the table below (Table 1.7).

Table 1.7 The ethylene sensitivity horticultural flowers (Smith et al., 2009).

Low sensitivity	Moderate sensitivity	High sensitivity
Tulip Daffodil	Lily Freesia Alstroemeria Anemone Dahlia Agapanthus	Carnation Geraldton Wax flower

Additionally, Kumar et al. (2008) claimed that there are three types of sensitive flowers. The first type is the ethylene production increasing with aging or following pollination can be caused by the senescence, such as carnation and petunia (Scariot et al., 2014; Serek et al., 1995). Second, the flowers will become sensitive to ethylene, which was produced during the pollinating process (e.g. cyclamen) (Halevy et al., 1984). The last type, the flowers so not elevate ethylene, but they are sensitive to the ethylene upon flower bud opening, for example, rose (Kumar et al., 2008; Serek et al., 1995).

1.3 Known ethylene effects on produce quality

1.3.1 Ethylene effects on colour change

Customers equate the appearance of horticultural products with quality (Saltveit, 1999). Ethylene accelerates chlorophyll degradation, converting green colour into yellow or orange colour, such as citrus and lemon (Saltveit, 1999; Sdiri et al., 2017). Both endogenous and exogenous ethylene also promotes senescence which causes the loss of green skin colouration in the supply chain. Banana's turn yellow in colour as influenced by ethylene, as the chlorophyll degrades (Janssen et al., 2013; Saltveit, 1999). Additionally, Wills et al. (2014) claimed that the green of bananas doubled when ethylene increased from 100 nL L^{-1} to 10 nL L^{-1}. Similarly, pepper (*Capsicum annuum*) can be completely coloured with the application of 2×10^6 nL L^{-1} ethylene (Graifenberg & Giustiniani, 1980). Mayuoni et al. (2011) also claimed that the ethylene can cause the degreening of chlorophyll and the accumulation of yellowing of citrus peel tissue. Similarly, it can cause the determined effects on green vegetables as well. Ethylene induce the yellowing of green vegetables, such as broccoli, bok choy and parsley has been estimated in many studies (Tian et al., 1995; Sledz et al., 2016; Cai et al., 2019; Wu et al., 2019). Therefore, ethylene is a choice within a high commercial value market to enhance the colours of products. Moreover, the removal of ethylene can delay colour changes. One example is that the papayas without applying ethylene colour changes slightly compared to those with 10^5 nL/L^{-1} ethylene application (Saltveit, 1999).

Ethylene can cause the colour discoloration as well. For example, Manjunatha et al. (2012) demonstrated that the ethylene can cause the browning in apples and Logan fruit (*Dimocarpus longa*). Similarly, ethylene can enhance the browning in table grapes has been studied as well (Kaplunov et al., 2015).

1.3.2 Ethylene effects on Firmness and Texture

Effects on texture has been found in many horticultural products, including sweet potato, cucumber, asparagus, and strawberry (Janssen et al., 2013; Saltveit, 1999).

Exposure to ethylene for a long time can cause tissue softening during fruit ripening which can progress into senescence (Iqbal et al., 2017). Some studies demonstrated that ethylene can induce the changes in ripening-related quality attributes of horticultural products effectively, including softening (e.g. avocado, kiwifruit, and apple), peel colour (avocado and apple), aromatic volatiles (e.g. apple, citrus and mango) and soluble solids (e.g. kiwifruit and apple) (Johnston et al., 2009; Sdiri et al., 2017; Gwanpua et al., 2018). Saltveit (1999) demonstrated that excessive exposure of ethylene can cause cucumbers and peppers to lose their crisp texture. Furthermore, the loss of firmness of peach was caused by ethylene has been reported as well (Manjunatha et al., 2012). Similarly, the melty texture can be caused by the improper treatment of ethylene (Zhang et al., 2017). A low level of ethylene can also affect the firmness of horticultural products undesirably. One research study estimated that 30 nL L^{-1} ethylene already can induce over softening in kiwifruit, as kiwifruit is quite sensitive to environmental ethylene (Saltveit, 1999). Yamaguchi et al. (1977) found that the softness honeydew melon can be enhanced by the application of ethylene (add the concentration). Moreover, Makkumrai et al. (2014) claimed that 10^5 nL L^{-1} ethylene can cause the stimulation of ripening in pears, and it can develop the mealy texture of pears.

Ethylene not only can cause the softening of products; it can also cause the toughening as well (Saltveit, 1999). Lipton (1990) estimated that excessive ethylene can stimulate the phenylpropanoid metabolism, phenolic compounds and lignification of tissue in asparagus. Furthermore, an undesirable level of ethylene can cause sweet potatoes to become hard to cook (Saltveit, 1999).

1.3.3 Ethylene effects on flowers

Ethylene can have negative effects on flowers too. Many research articles demonstrate that the petal abscission, petal wilting, and senescence can be caused by ethylene (Faragher & Mayak, 1984; Reid & Wu, 1992; Ichimura et al., 1998; Eouter & Doorn, 2002). In Wouter and Doorn (2002), 300 species of flowers were treated with 300 nL L^{-1} ethylene. Flower abscission was found to be very sensitive to ethylene in all species except *Cymbidium*. Similarly, another study did the ethylene treatment to *Eustoma* flowers and they claimed that the senescence was accumulated at 1000 nL L^{-1}, 6 days after anthesis. Similarly, Woltering and Doorn (2002) also studied in the ethylene impact with 93 species of 23 families, and they found most flower species, except *Campanulaceae*, *Caryophyllaceae*, *Malvaceae*, and most *Orchidasea*. Woltering and Doorn (1988) found that wilting was the primary symptoms of senescence. Membrane permeability and senescence were increased with the appearance of ethylene in cut rose flowers (Farahjer & Mayak, 1984). The major symptoms and the sensitivity of flowers are shown in Table 1.8. In this table, plants have different classes: Class 0 (insensitive); Class 1 (upon 33% reduction of the vase life); Class 2 (33%-66% reduction of the vase life); Class3 (66%-99% reduction in vase life) and Class 4 (immediate drastic effect) (Shahri & Tahir, 2011).

Table 1.8 The differences of symptoms and sensitivity in different flowers (A: Abscission, W: Wilting; WA: Wilting followed by abscission) (Shahri & Tahir, 2011).

Family	Symptoms	Sensitivity
Monocotyledonae		
Amaryllidaceae	W/WA	0-3
Iridaceae	W/A	0-4
Liliaceae	W/A	0-3
Orchidaceae	W	3-4
Dicotyledonae		
Campanulaceae	W	2-4
Caryophyllaceae	W	4
Compositae	W	0-1
Dipsaceae	W/WA	2-3
Euphorbiaceae	W	1
Geraniaceae	A	4
Labiatae	A	4
Malvaceae	W	4
Primulaceae	W	2-4
Ranunculaceae	A/AW	0-4
Rosaceae	A	3-4
Scrophulariaceae	A	3-4
Umbelliferae	W	0

1.4 Ethylene within the supply chain

1.4.1 Ethylene management

Ambient atmospheric conditions in the supply chain highly influence the resulting quality of horticultural goods. Fruits and vegetables are perishable. Losses can contribute to 40 - 50% of global waste every year (FAO, 2015). Appropriate conditions can maintain the freshness of transported and stored perishable goods. Use of refrigerated containers or transport equipment or cool stores is appropriate (Salveit, 1999; Martinez-Romero et al., 2007; Zhang et al., 2017). Controlling atmospheric conditions in these environments is key to maintaining freshness and extending the shelf life of horticultural products postharvest. Numerous studies have demonstrated that ethylene is one of the most important gases to be controlled and monitored in the fruits and vegetables (Argueso et al., 2007; Janssen et al., 2013). Ethylene management can be a significant measure to reduce fresh produce waste along the supply chain (Blanke, 2015). Many research studies found that ethylene plays an important role in the fruit supply chain and has effects on many events such as seed germination, senescence, abscission, cell elongation and fruit ripening (Ella et al., 2003; Binder, 2008).

1.4.2 Ethylene concentration in the supply chain

With the increasing interest in the ethylene management, some studies have focused on the ethylene concentration in different handling areas. Pathak et al. (2017) demonstrated that higher than normal ethylene concentration (1-5 nL L^{-1}) could be found in the vicinity of storage facility of fresh produce, due to ethylene being produced by climacteric fruits and the movements of trucks, tractors, and forklifts during the supply chain (Warton et al., 2000; Morgott, 2015). Different areas in the supply chain may have different concentrations of ethylene. Warton et al. (2000) estimated that the ethylene concentration at supermarkets ranged from 17 - 35 nL L^{-1}, while the wholesale markets and distribution centre can be around 60 nL L^{-1}. One study stated that the ethylene can be build up to around 50 nL L^{-1} with combustion products (Keller et al., 2013). Moreover, 10 - 100 times ambient ethylene concentration can be accumulated in some cases, such as heavy traffic, greenhouses and packages (Keller et al., 2013). Similarly, Rees et al. (2011) found ethylene concentrations in retail store of 50 and 3600 nL L^{-1} in storage facilities.

1.4.3 Ethylene effects in the supply chain

Keeping a safe level of ethylene is quite important in the market system, although it seems that there are still some conflicts on the accurate threshold level of ethylene in the postharvest environment of fresh produce. Wills et al. (2000) estimated that 100 nL L^{-1} can be the threshold level of ethylene as this level is unlikely to induce any undesirable physiological effects on horticultural products. Moreover, another study suggests that when the level of ethylene can be reduced from 100 nL L^{-1} to < 5 nL L^{-1} the postharvest life can be extended for around 30 types of fruits and vegetables (Wills et al., 2000). However, others demonstrated that a wide range of non-climacteric produce was adversely affected when the concentrations of ethylene were below 100 nL L^{-1} (Willis & Wang, 1996; Willis et al., 1999). Similarly, Peacock (1972) found that the 100 nL L^{-1} cannot be a safe threshold level for banana, possibly because banana can produce ethylene by itself. Also, this article is quite out of date, hence the finding of this study may have different results. All the findings showed that it is hard to accurately conclude on a 'safe level of ethylene' as different horticultural products will have a different response to different levels of ethylene. Nonetheless, most studies claim that low, medium and high rates of ethylene are considered to be ≤15 nL L^{-1}, 15 -100 nL L^{-1}, and 100 nL L^{-1}respectively.

Table 1.9 The ethylene levels in handling areas which were rated as low, medium or high (Wills et al., 2000).

Situation	n	Low (≤15 nL L^{-1})	Medium (>15 - <100 nL L^{-1})	High (≥100 nL L^{-1})
		Wholesale market		
Air	36	5	78	17
Non-climacteric	389	10	74	16
		Distribution centre		
Air	13	0	85	15
Storage room	35	0	40	60
		Supermarket retail stores		
Receival	49	47	53	0
Storage	49	8	90	2
Display	49	39	61	0
		Consumer market		
Refrigerator	30	17	53	30

Nowadays, some studies focus on measuring ethylene levels in different storage environments to figure out the optimal atmospheric condition for prolonging postharvest storage life of fresh produce. For example, Wills et al. (2000) compared different levels of ethylene in different handling areas (Table 1.9). The authors define 15 nL L^{-1} as the acceptable low level of ethylene, while ≥100 nL L^{-1} is unacceptable. Results suggest that 15%-17% of the wholesale markets had high levels of atmospheric ethylene, while 5-10% were kept at a low level. However, the ethylene concentration in the distribution centre was always medium to high and the storage area was never at low level. It is likely that horticultural products stored at the distribution centre, especially in the storage rooms, can be exposed to higher risks of shelf life loss. Wills et al. (2000) estimated that the loss of postharvest life in the wholesale market and distribution room was substantial (25%-30%). On the contrary, the supermarket retail stores can be the most suitable environment as the mean ethylene level was low at 25 nL L^{-1} and the average loss of postharvest life was 15%. Meanwhile, the ethylene level in domestic refrigerators was found to be quite high at 3×10^4 nL L^{-1}. Therefore, the authors claim that the ethylene level in the supermarket retail store is much more favourable.

1.4.4 Ethylene management in the supply chain

It may be necessary to specifically manage ethylene to extend the postharvest life of products. Wills et al., (2015) found that there were 10% and 30% potential loss of products, when products were stored in 15 or 100 nL L^{-1} respectively. Rees et al. (2011) compared the products stored below 5 and 100 nL L^{-1} and demonstrated that a 60% extension of postharvest storage life can be achieved when stored below 5 nL L^{-1} ethylene.

In order to minimise the adverse effects of ethylene on the horticultural products, the inhibition of biosynthesis and receptors level, and the use of ethylene oxidizer should be achieved. There are several treatments to minimise ethylene damage which will be discussed in this chapter, including CO_2, 1-methylcyclopropene (1-MCP) and potassium permanganate. It is recommended to separate the products, based on the ethylene sensitivity (Watkins, 2016). However, within the supply chain, ethylene sensitive products are often kept with ethylene producing products due to the high cost of creating separate facilities (Keller et al., 2013). This introduces a risk of loss of postharvest life considering that 10 nL L^{-1} can induce detrimental changes for some products (Wills et al., 2015). As a gas, ethylene can diffuse easily from one item to another, as it has same specific mass as air (Blanke, 2014). Lawton (1999) found that for refrigerated containers loaded with kiwifruit and apple, the ethylene concentration can be 1 - 8 nL L^{-1} and 500 - 15000 nL L^{-1} respectively. Moreover, Keller et al. (2013) found that the ethylene concentration of containers with apples, pears and grapes was around 50000 nL L^{-1} ethylene, but apples and pears were the main source of ethylene.

1.5 Broccoli

The second component of this study investigates the effects of environmental ethylene on broccoli storage. For this reason, a review of the postharvest knowledge of broccoli is provided below.

1.5.1 Quality and Postharvest Physiology of Broccoli

Broccoli (*Brassica oleracea* var. *italica*) is a common and popular vegetable in the market, containing a wide range of phytochemicals, including gluosinolates, flavonols, carotenoids, vitamins and sugars (Tian et al., 1995; Albe, et al., 2003; Soyasal, 2004; Jones, et al., 2006; Kaiser et al., 2012; Pan & Sasanatayart, 2016). Broccoli is high in vitamin C, rich in soluble fibre and nutrients (Cai et al., 2019). Broccoli belongs to the *Cruciferae* or *Brassicaceae* family and is a major food crop globally. Increasingly more people show an interest in broccoli, as it is associated with the dropped rate of rectal cancers and some disease of the stomach, colon and lung (Jones et al., 2006).

Broccoli is considered short shelf-life with high ethylene sensitivity (Tian et al., 1995; Tian et al., 1996; Lu, 2007). Yellowing through degradation of chlorophyll and weight loss are quality parameter to lead the short shelf-life of Broccoli (Gong & Mattheis, 2003; Cefola et al., 2010).

Gong and Mattheis (2003) claimed that the green colour is the quality index, as the degreening of broccoli can occur rapidly after harvest, limiting postharvest storage (Cai et al., 2019). Other quality reduction processing includes loss of turgor, nutrients (e.g. sugars and vitamin C) (Jones et al., 2006). Quality loss and the short shelf can limit the presence of broccoli in the market.

1.5.2 Quality loss of broccoli

1.5.2.1 Colour change

Colour is a quality attribute that influences consumer acceptability during shelf life (Kaiser et al., 2012). Moreover, colour can pose a primary role in the perception of sweetness and pleasantness (Wu et al., 2019). Berset and Caniaux (1983) illustrated that the colour is nonuniformly distributed over the surface of the food. Chlorophyll is the major predominant pigment of green plants. The colour changes and chlorophyll degradation of broccoli can be induced via ethylene rapidly and have been reported by several studies (Forney, 1995; Tian et al., 1995; Tian et al., 1996; Jones et al., 2006). Chlorophyll degradation can be used as an index to evaluate the senescence of green plants (Wu et al., 2019). Many studies assess that the quality of broccoli with respects to shelf life as influenced by colour chlorophyll and weight loss (Asoda et al., 2009; Cai et al., 2019).

Bansal & Aggarwal (2011) claimed that each region of the image can be characterized via feature (e.g. intensity and colour) via the colour model. Many studies use the CIELab colour space to assess the colour quality of green vegetables (Bai et al., 2013). The colour space coordinates a white reference (Simonot et al., 2011). Also, the CIE spaces are based on Munsell colour system and it can focus on the relationship between of three attributes – Munsell hue, Munsell chroma, and Munsell value (Mahyar et al., 2010). Some studies presented only h value information as colour and the angle between 0° and hypotenuse, less studies provide information on the changes in C and L value, as C represent (degree of departure from grey to pure) and L was lightness from black to white (McGuire, 1992; Sanmartín et al., 2012). Moreover, chroma C can be calculated as $(a^2+b^2)^{1/2}$ (McGuire, 1992).

Overall, there was an increasing trend of chroma (C value) in the first 4 storage days of banana fruits with continuous ethylene application, and 10^4 nL L^{-1} treatment had the highest C value (around 50) at the fourth day than other treatments, while the control group had the lowest (Gutierrez-Martinez et al., 2010). Fan and Mattheis (2000) measured C value broccoli during 18 days with 4 treatments (control, 1000 nL L^{-1} MCP, 1000 nL L^{-1} ethylene and MCP + Ethylene) and they found the chroma increased during the storage time and ethylene treatment had the highest C value, which was around 28 at the last day.

There is a decreasing trend of hue angle (h value) during the postharvest process of broccoli. Tian et al. (1995) measured h value of three kinds of broccoli (Shogun, Green belt and Green beauty) and they demonstrated there was a negative relationship between the ethylene accumulation and h value, as the Green belt had the highest ethylene production (4 nL L^{-1}) and lowest h value than others. Similarly, another study treated broccoli with 5000 nL L^{-1} ethylene, they estimated that the ethylene decreased during the increase of storage time and the broccoli with ethylene had lower h value than the control group (Asoda et al., 2009). Gong and Mattheis (2003) estimated that the h value of broccoli with 10^6 nL L^{-1} dropped faster than the control group.

There are less studies analyse the lightness (L value) of ethylene treatment. Gutierrez-Martinez et al. (2010) they found the l values of banana fruits with different treatments at the 10th day was similar with the first day. It seems that the 10^4 nL L^{-1}treatment (L=55 - 56) was similar to 10^5 nL L^{-1} and 10^6 nL L^{-1} (L=62), but they were higher with the nontreated banana fruits (L=51).

A higher redness/greenness (a value) and yellowness/blueness (b value) (or lower h values) describes yellowing of vegetables, corresponding to the lower chlorophyll content (Fang et al., 2016; Wu et al., 2019). Gong & Mattheis, (2003) found the broccolis treated with 10^6 nL L^{-1} had a higher b value than the control group. However, they found 10^6 nL L^{-1} ethylene treatment had the lower a value than the control group (Gong & Mattheis, 2003).

1.5.2.2 Weight loss

Weight loss can cause shrivel and the weight loss during the storage of products (Lu, 2007). Sabir (2012) claimed that Broccoli has a short shelf life due to yellowing, water loss and decay. Similarly, Serrano et al. (2006) found that the broccoli florets stored for 20 days with 1℃ and 90% RH can have a high weight loss (46.36 ± 1.04%) during the storage and this phenomenon affects the marketability. Also, Broccoli with 20% weight loss has been found in 5 days with room temperature (Serrano et al., 2006). This can be accounted for the losses of compounds, such as flavonoids, glucosinolate and vitamin C (Vallejo et al., 2003).

Weight loss is an important index to assess the quality, as the tissue dehydration and the increase of elasticity and fibrous can increase during the storage

(Serrano et al., 2006; Gomes et al., 2008). Similarly, Huang and Liang (2012) also claimed that weight loss and profile reduction can cause the reduction of marketability of horticultural products. Moreover, the weight loss with loss of glucoraphanin, precursor, 4-methylsulfinyl butyl isothiocyanate (sulforaphane) can be found during the storage (Rangkadilok et al., 2002; Duarte-Sierra et al., 2017).

1.5.2.3 Decay

Winkler et al. (2007) estimated that the potential decay symptom can be detected after 28 days at 4℃ and the decay can be affected by the combination of the storage time and temperature. Additionally, soft rot can be caused by the infection of *Pectobacterium* and *Pseudomonas* (Gašić et al., 2014). Gašić et al. (2014) also claimed that the water-soaked area can be found from the stem area to the entire plant, which progressed the soft rot decay Similarly, Huang and Jiang (2012) demonstrated that severe rotting can be caused by fungi and bacteria, which induce the loss during storage.

1.5.3 Postharvest physiology of broccoli

Broccoli is a rapidly developing floral vegetable, where the florets senescing rapidly after harvest being one of the most obvious features (Tian et al., 1994; Tian et al., 1995). This rapid postharvest senescence is due to the floral head being separated from nutrients, hormones and energy, which were provided by the roots and leaves at harvest (Tian et al., 1995). As a consequence, the shelf life of broccoli is quite short, approximately only 2-3 days at 20 °C (Jacobsson et al., 2004). Hansen et al. (2000) claimed that the temperature can be the factor to affect the senescence rate of broccoli. It is easier to induce the quality loss of broccoli via high storage temperature (Winkler et al., 2007) For example, broccoli can store around 7 days with 10 °C that was reported via Hansen et al. (2001)'s study, while Makhlouf et al. (1989) estimated broccoli can be stored around 31 days at 0 - 1°C. Besides, another study found that the broccoli can be stored 3-4 weeks at 0 °C, while only 2-3 days at 20 °C (Jacobsson et al., 2004).

Respiration rate contributes the metabolism and it can maintain the activities and physio-chemical reactions in the horticultural products (Taiz & Zeiger, 2009). There is a relationship between senescence and respiration, as the senescence is associated with low energy and a high respiration rate (Li et al., 2016; Jiang et al., 2017). It is known that extending shelf life and delaying senescence can be achieved by controlled and modified atmosphere (Kato et al., 2002). Lower O_2 and higher CO_2 can benefit the storage of broccoli (Izumi et al., 1996). Hansen et al. (2000) claimed that 1-3% O_2 and 5 – 10% CO_2 was recommended to slow down the senescence process. Li et al. (2016) found that 50% O_2 + 50% CO_2 can reduce 36.57% respiration rate and was a potential method to extend the shelf of broccoli. Furthermore, 10% O_2 + 5% CO_2 and 5% O_2 + 95% N_2 can reduce 13 % and 21% respiration rate respectively.

Table 1.10 Previous studies about ethylene treatments on broccoli.

Treatment	Environment conditions	Comments	Reference
Air (control); 1000 nL L^{-1} ethylene (C2H4); 12 h 1000 nL L^{-1} – (MCP); MCP + C2H4	10℃	The colour changes were detected after 2 days and the ethylene treatment had the lowest hue angle (around 122) and highest chroma (around 29). The yellowing can be reduced by MCP	Fan & Mattheis, 2000
Air (control); 5000 nL L^{-1} Ethylene; 40 pmolmL^{-1} Ethanol; Ethanol + Ethylene	20℃	The yellowing and ethylene biosynthetic enzymes can be induced (*BO-ACO1* and *BO-ACO2*). The ripening and senescence can be reduced and inhibited by the ethanol.	Asoda et al., 2009
Air (control); 10^5 nL L^{-1} Ethylene; 1000 nL L^{-1} 1-MCP; 1-MCP + Ethylene	5℃	The severe yellowing, chlorophyll degradation and complete flowering can be caused via the ethylene treatment, while 1-MCP can inhibit the ethylene-induced decline effectively.	Cefola et al., 2010
10^5 nL L^{-1} Ethylene	10℃	The yellowing increased after 2 days with ethylene treatment, and it also causes the enzyme activity and expression of genes encoding key enzymes also was investigated. The degradation of chlorophyll and carotenoids.	Cai et al., 2019

Ethylene plays an important role in plant maturation and colour development (Cai et al., 2019) and an important role in the loss of chlorophyll (Gong & Mattheis, 2003). Tian et al. (2014) found that ethylene from either endogenous production or exogenous application stimulated chlorophyll loss and promoted the yellowing process of harvested broccoli florets. Asoda et al. (2009) demonstrated that the ethylene can accumulate the yellowing of broccoli, as the colour score dropped with 5000 nL L^{-1} ethylene fastest than other treatments (air treatment, ethanol treatment, and ethylene + ethanol treatment) and the broccoli can become 100% yellowing at the end of the trial. Rees et al. (2011) claimed that broccoli can be sensitive to <100 nL L^{-1} ethylene. The broccoli treated with ethylene has been studied in the past, but there is only a few information about the effects of different ethylene treatment on broccoli (Table 1.10).

Treatment of broccoli with ethylene inhibitors, such as 1-MCP (1-methylcyclopropene) and aminoethoxyvinylglycine acid (AVG) can also delay the senescence and the chlorophyll loss in broccoli (Tian et al., 1994; Forney et al., 2003).

1.6 Conclusion and Research Objectives

Ethylene has the potential to cause deleterious effects on fresh. However, ethylene concentrations in the environment will differ both with location and time. There are only a few articles that measuring ethylene in a real fresh produce chain. As a result, there is insufficient information about the expected approximate ethylene concentrations in different parts of the supply chain such as handling areas, supermarkets and distribution centres. Contemporary ethylene detecting techniques, such as the MACView can be used in the commercial area and enable collection of temporal data at lower ethylene concentrations (nL L^{-1}) than before.

Questions that are remain difficult to answer include:

- *What is the ethylene concentration within the fresh produce chain in different handling areas?*
- *How does the ethylene concentration change during the day in these different locations?*
- *Given the ethylene concentrations measured are they likely to have impact the quality of common crops?*

(a) Given these questions two small independent research projects were conducted. In order to gain greater understanding of likely ethylene contamination in the supply chain, surveys of ethylene concentrations in commercial facilities were conducted. Given these results, ethylene concentrations in the ranges measured were applied to broccoli as a case study, in order to study the potential quality impacts of these environmental ethylene concentrations. To analyse the relationship between the horticultural products, display method and ethylene concentration

(b) To figure out the main ethylene level in common handling areas, such as supermarkets, flower store, and distribution centres.

(c) To investigate the main changes of the ethylene concentration in the same locations during the days

(d) To determine the shelf life of some common horticultural products

(e) To determine the influences that are caused by the common ethylene level to the shelf life of the products.

1.7 Relevance of the study

This research is a benefit to the fresh produce chain, and it gives suggestions to the distribution centres, supermarkets, and flower stores, which are related to control the ethylene level or manage the product display method to extend the shelf life of the products in some ways. These findings can provide the information to further studies on how to reduce the detrimental impacts in different areas.

Chapter 2 Industry Environment Survey

2.1 Introduction

The recognition of the significance of controlling ethylene in the fresh produce chain is growing, given the effect on product quality. It is prudent to ensure ethylene concentrations in the fresh produce chain remain below critical concentrations that induce substantial product deterioration. Knowledge of the ethylene concentration enables management and control in order to maximise the shelf life of products.

Environmental ethylene in the supply chain can come from a number of sources. Industrial activity is usually accompanied by high use of combustion engines that create ethylene when combustion is incomplete. Sawada et al. (1989) found that burning exhausts can produce ethylene. One study measured the ethylene concentration in Washington city and they found the air in city centre had much higher ethylene level (700 nL L^{-1}) than the areas outside the circumferential beltway (39 nL L^{-1}) due to the automobile exhaust (Abeles & Heggestad 1973). The ethylene from flue gas in Long Island, New York was up to 1.5×10^3 nL L^{-1} (Luria et al., 2000). Similarly, Morgott (2015) reported that the ethylene concentration with air pollution, such as petrol exhaust and smoking, can range up to 5×10^4 nL L^{-1} – 10^5 nL L^{-1}.

Horticulture products also produces ethylene, each at a different rate. For non-climacteric products, 100 nL kg^{-1} ethylene can be produced (Kader, 1980). By comparison, 103 - 104 nL kg^{-1} can be produced in the preclimacteric period, while the ethylene concentration increased by at least 10-fold when the product ripens (Keller et al., 2013). The ethylene production of some major products is shown in Table 2.1.

The ethylene concentration in the supply chain is reported by several studies. Keller et al. (2013) demonstrated that the ethylene concentration in ambient area is usually below 50 nL L^{-1}. However, the ethylene concentration can be up to 50 nL L^{-1} with petrol exhaust machine. For example, heavy traffic can increase 1000 nL L^{-1} even more (Keller et al., 2013). One study found the ethylene concentration in distribution centre ranged from 72 nL L^{-1} - 548 nL L^{-1}, while ethylene concentration in the coolroom with apples was 3612 nL L^{-1} (Rees et al., 2011). Furthermore, the ethylene concentration in supermarkets is usually between 0 nL L^{-1} - 100 nL L^{-1} (Wills et al., 2001; Rees et al., 2011). Wills et al. (2011) monitored 49 supermarkets and found only 2 supermarkets had environmental ethylene concentrations of more than 100 nL L^{-1}.

Table 2.1 Ethylene production rate of some fresh (Rees et al., 2011; Keller et al., 2013).

Fresh produce type	Ethylene production rate nL kg^{-1}
Cut flowers	Very low < 10^2
Broccoli	Very low < 10^2
Cabbage	Very low < 10^2
Carrot	Very low < 10^2
Cherry	Very low < 10^2
Grape	Very low < 10^2
Mushroom	Very low < 10^2
Lettuce	Very low < 10^2
Onion	Very low < 10^2
Orange	Very low < 10^2
Potato	Very low < 10^2
Cucumber	Low 10^2 - 10^3
Kiwifruit	Low 10^2 - 10^3
Peach	Moderate 10^3 - 10^4
Pear	Moderate 10^3 - 10^4
Banana	Moderate 10^3 - 10^4
Avocado	Moderate 10^3 - 10^4
Apricot	High > 10^4 - 10^5
Apple	Very High > 10^4
Passion fruit	Very High > 10^4

In this study, the ethylene concentrations of different locations within local supply chains were measured in a survey fashion to assess whether typical ethylene environments may represent a risk to storage life and produce losses. This quantitative research adds to the paucity of data on ethylene concentration in commercial handling areas. With this data, likelihoods of damage to products as a result of exposure to ethylene can be assessed.

2.2 Materials and methods

2.2.1 Project Overview

The experiment consisted of ethylene monitoring in three different types of supply chains, including a distribution centre, several supermarkets and a flower distributor (Table 2.2). All sites were located in the Manawatu area of New Zealand and were monitored on condition of anonymity.

Table 2.2 The overview timeline and information of the project.

Location	Timing	Locations measured
Distribution Centre	13th May – 10th June	6
Supermarket A	17th July – 6th August	3
Supermarket B	7th August – 4th September	4
Supermarket C	1st November – 3rd December	3
Supermarket D	9th January – 7th February	1
Flower distributor	3rd September – 28th September	4

2.2.2 Ethylene monitoring system

The portable ethylene detector, Analyser of EMS (MACView, Netherlands), was used to monitor the ethylene concentrations. This equipment features an analyser system with an integrated control mechanism for reliable ethylene concentration measurement (Figure 2.1). The equipment has a lot of advantages, including convenient to use and transport, high sensitivity (Shekarriz & Allen, 2008; Blank & Shekarriz, 2012; Janssen et al., 2013). The range of the ethylene measurement is 0-5000 nL L^{-1} (Verschoor,2017).

Figure 2.1 The MACView in postharvest lab at Massey University.

The MACView is the electrochemical sensor (EC) and has been used previously in commercial situations to monitor ethylene in postharvest storage areas (Boerman et al., 2016; Lerud et al., 2019). The EC sensor consists of two electrodes, which are made via platinum or gold nanoparticles, held at a fixed voltage (a porous substrate) (Shekarriz et al., 2008; Zevenbergen et al., 2011). Once the air goes into the sensor, it can be oxidized on the interface of an electrode (Shekarriz & Allen, 2008). This process will follow the formulation below (Janssen et al., 2013):

$$C_2H_4+3O_2 => 2CO_2+2H_2O$$

After the oxidation process, the whole electronic circuit is completed and the ethylene concentration can be obtained (Janssen et al., 2013; Lerud et al., 2019).

Moreover, it seems that the MACView can utilise an onboard ethylene gas cylinder for a calibration standard to provide a periodic span correction (Lerud et al., 2019). In the current study, the calibration of the MACView was conducted before getting measurements in the supply chain. MACView was calibrated against gas mixes with known ethylene concentrations. The calibration ethylene concentrations were achieved by using two mass flow controllers. Mass flow controllers can ensure the flow and its associated gas into the detector via predetermined rate (Doyler, 1987). In the current study, one mass flow controller was used to control the air and the other to control ethylene. The total flow rate was set as 300 mL/min and the 4500 nL L^{-1} ethylene standard was used at different flow rates to make up gas mixes of various ethylene concentrations (45 nL L^{-1}, 92 nL L^{-1}, 502 nL L^{-1}, 960 nL L^{-1} and 4500 nL L^{-1}). The following calculation was used for calibration:

Ethylene concentration (nL L^{-1}) = Flow rate of ethylene (ml/mins)/ Total flow rate × concentration of ethylene standard

During in-field measurements, the MACView was provided with a constant power supply to avoid interruption of measurements due to battery exhaustion. It is important that the device should be stabilized when in use. Real-time data were updated in a web-based portal (*http://www.mymacview.com*) and constantly monitored to ensure consistent data capture.

The measurement of oxygen, carbon dioxide and ethylene, was tested by Wageningen Food & Biobased Research (WFBR) in 2017, demonstrating the suitability of using MACView in horticultural settings (Verschoor, 2017). Moreover, MACView was applied to monitor the ethylene generation *in situ* of apples (Lerud et al., 2019). As there are limited studies reporting the use of MACView to measure ethylene concentrations in the supply chain, the current study could potentially provide useful information on the effectiveness of MACView for monitoring environmental ethylene.

For this study, the MACView was connected to a multiplexer, enabling sequential monitoring of different locations. The multiplexer was connected to the MACView via 6 mm nylon tubing (Leda-lon Nylon 12, Leda, New Zealand), which has two outlet branches. One branch fed into the MACView sampling port and the other was used for exhausting any excess gas and avoiding under or over pressure. Two different multiplexers (with 5 and 6 input points respectively) were used in this study. The tube (length: 5-25m) was run from each sampling location to the inputs of the multiplexer. All the tubes were firmly secured to the wall using tape.

During sampling, three readings of the atmospheric ethylene concentration from the same location were collected by MACView, at a speed of 15 mins per measurement (Verschoor, 2017). After this, the multiplexer switched to the next input connection point to enable sampling from a separate room/location. The sequence at which the rooms were sampled were pre-programmed and this allowed measurements from different locations in a pre-defined order. A general check was conducted once a week to ensure the device was operating properly and the tubes remained in the same location.

2.2.3 Other environmental monitoring

The temperature and relative humidity in different storage and/or handling areas were also monitored using I-Buttons (Maximum Integrated, San Jose, CA). I-buttons were placed in each location next to the inlet of the tube for ethylene monitoring. During this process, the tape held the back of I-buttons and the sensor point of I-buttons should be exposed to the air. Sample rate of the I-buttons was set to take an instantaneous measurement every 5 minutes. Prior to the survey, the I-Buttons were calibrated by RH% of KCl (Potassium chloride) saturated salt solution (58.86%, 75.65%, 87.67% and 96.27%).

2.2.4 Data analysis

The ethylene data in each location was analysed by constructing histograms and plots of relative frequency of measured ethylene concentrations. In order to study time effect on ethylene concentration, 24 h plots on each calendar day were also constructed. As MACView collects three readings per hour from each room, the average of three readings per hour was used to plot the 24h daily trend. All data was managed in Excel (64-bit Version, Microsoft, USA) and the figures were plotted by Sigmaplot (Version 14.0, Graph software, USA).

2.2.5 Location description

2.2.5.1 Distribution centre

There are 6 chiller rooms in the distribution centre that was surveyed (Figure 2.2). However, chiller room 1 was kept empty most of the time and it was also used for temporary storage of random products and hence its ethylene concentration was not measured. All the chiller room were the same size which was 4.35 m × 3.56 m × 3.40 m. Not every room had a strip curtain, with room 3 and 4 not having them. Whether the room has a plastic curtain, can be a factor that impacts the results, as the strip curtains can stop the air exchange from the room to the external environment to some extent. All the sampling points were positioned near the entrance of the chiller rooms and I-buttons were taped next to the sampling points. There were forklifts with petrol exhaust moving between storage areas and chiller rooms, which might affect the ethylene concentration.

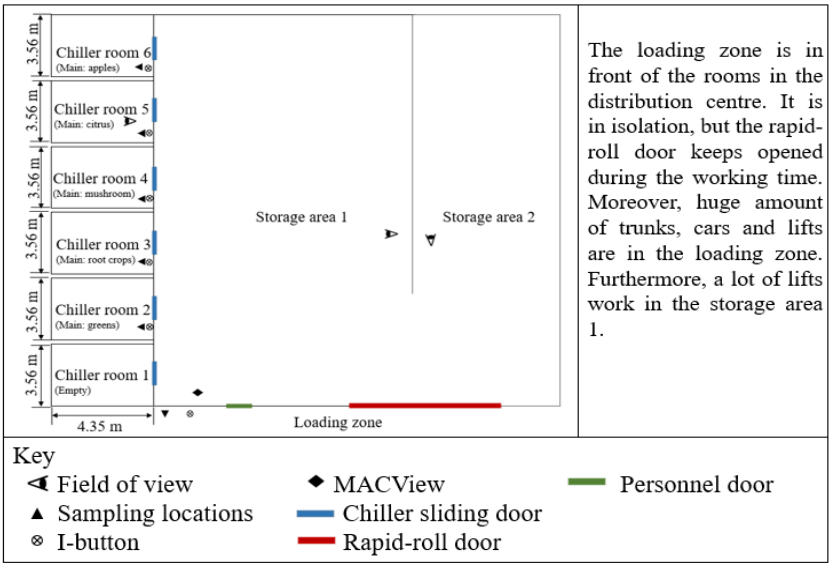


Figure 2.2 Plan of distribution centre.

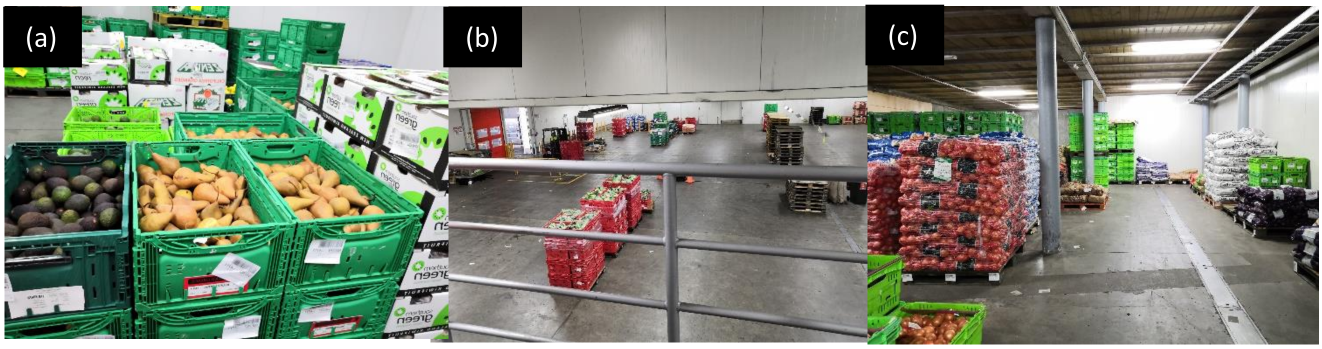


Figure 2.3 The picture of produce coolstore area (a) and the storage area 1 at room temperature (b) and the storage area 2 at room temperature (c) in distribution centre.

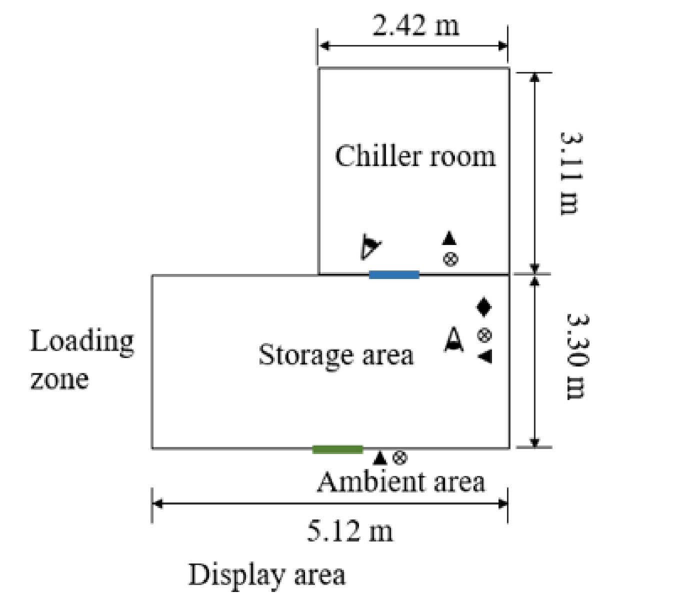
Different chiller rooms stored different crops (Table 2.3) which can cause the ethylene concentration differences between each room. For instance, chiller rooms 5 and 6 could have high ethylene concentrations due to the high production rates of avocado, peaches and apples.

Table 2.3 Details of coolstore rooms and produce inside for distribution centre.

Area	**Room size**	**Main products**	**Weight (kg)**	**Door**
Chiller room 2	4.35 m × 3.56 m × 3.40 m	Greens	275	With strip curtains
Chiller room 3	4.35m × 3.56m× 3.40 m	Root crops	480	Without strip curtains
Chiller room 4	4.35m × 3.56m× 3.40 m	Mushroom Red onions	46 72	Without strip curtains
Chiller room 5	4.35m × 3.56m× 3.40 m	Citrus Kiwifruit Avocado Peaches	750 132 88 165	With strip curtains
Chiller room 6	4.35m × 3.56m× 3.40 m	Apples	855	With strip curtains

2.2.5.2 Supermarket A

In supermarket A, ethylene concentration of three areas (ambient, storage and coolstore) were measured (Figure 2.4). The chiller room did not have a strip curtain to separate two different temperature zones, whereas the shelf area had a strip curtain to separate the air between outside and the storage area. Some forklifts with petrol exhausts were observed to park in the loading zone next to the storage area, which might cause higher ethylene concentrations throughout the day when in use.



The display area of supermarket A is in front of the rooms, while loading zone is on the left side of the rooms. The display area is in isolation, while the loading zone is not. Moreover, many trunks, cars and lifts are in the loading zone.

Key
- ◄ Field of view
- ▲ Sampling locations
- ⊗ I-button
- ◆ MACView
- Chiller sliding door (blue line)
- Personnel door (green line)

Figure 2.4 Plan of the storage areas in supermarket A.

Figure 2.5 The picture of produce chiller area (a) and the storage area with room temperature (b) in supermarket A.

The chiller room contained high ethylene producing produce such as apples, citrus, kiwifruit and banana were mainly stored and placed in the chiller room to keep fresh, while the storage area had a mix of low and high ethylene producing crops such as avocado, tomatoes, pears and potatoes.

Table 2.4 Details of room and product inside for supermarket A.

Areas	**Room size**	**Main products**	**Weight (kg)**	**Door**
Chiller room	2.42m × 3.11m	Apples	190	Without strip curtains
		Citrus	180	
		Kiwifruit	23.1	
		Banana	72	
Storage area	5.12m × 3.30m	Avocado	88	With strip curtains
		Tomatoes	40	
		Pears	80	
		Potatoes	59.4	

2.2.5.3 Supermarket B

The storage area of this supermarket consisted of shelf area, one chiller room, banana area, storage area and outside (Figure 2.6). The banana area was chosen as banana can produce high levels of ethylene and this area in the supermarket B occupied quite a large size.

There were 2 sampling points in the chiller room and 1 each in the storage, ambient and banana areas.

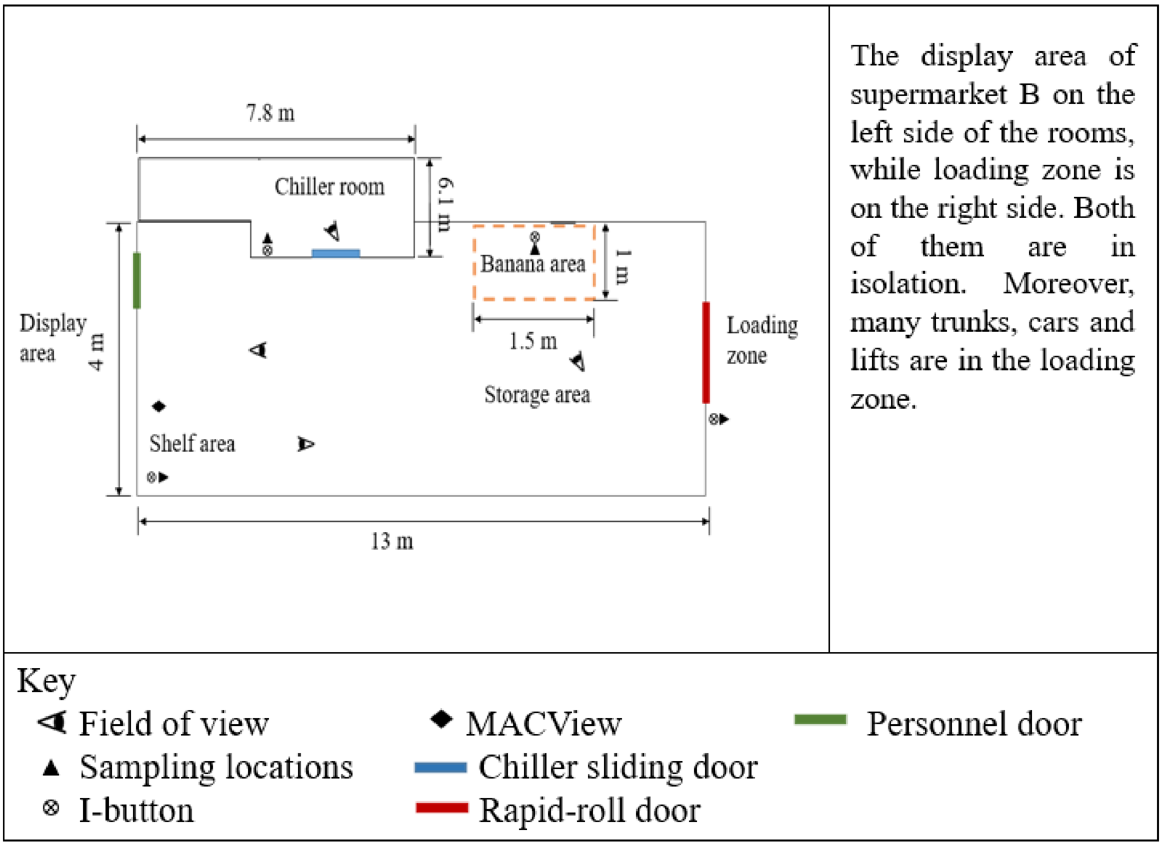


Figure 2.6 Plan of the Supermarket B.

Figure 2.7 The picture of produce chiller area (a), the banana area with room temperature (b), the storage area (c) in supermarket B.

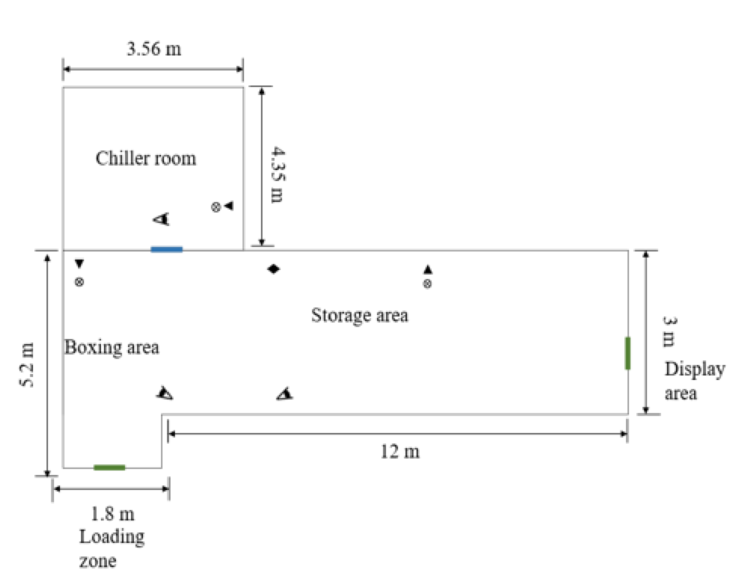
The chiller room contained a mix of products that are highly sensitive to ethylene such as kiwifruit with products that have high ethylene production rates such as apples.

Table 2.5 Details of room and product inside for supermarket B.

Areas	**Room size**	**Main products**	**Weight (kg)**	**Door**
Chiller room	5.20 m × 2.60 m × 2.70 m 2.60 m × 3.50 m × 2.70 m	Apples Citrus Kiwifruit	950 645 66	With strip curtains
Banana area	1.50 m × 1.00 m × 2.70 m	Banana	540	Without strip curtains
Shelf area	13.00 m × 4.00 m × 2.70 m	Potatoes	297	With strip curtains

2.2.5.4 Supermarket C

Figure 2.8 illustrates the setup of the ethylene monitoring system. Three locations, specifically the chiller room, display area and ambient area were monitored (Figure 2.8. and Figure 2.9.). In addition, the bakery was stored inside the chiller room as well. This factor should be taken into consideration during analysing the result.



The display area of supermarket C is on the right side of the rooms, while loading zone is in front of the boxing area. The display area is in isolation, while the loading zone is not. Moreover, many trunks, cars and lifts are in the loading zone.

Key
- ◀ Field of view
- ▲ Sampling locations
- ⊗ I-button
- ◆ MACView
- Chiller sliding door (blue line)
- Personnel door (green line)

Figure 2.8 Plan of supermarket C.

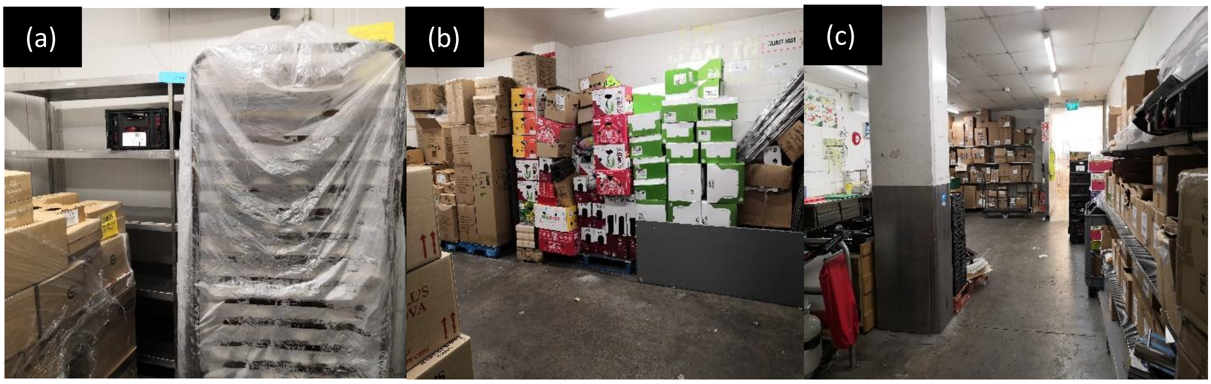


Figure 2.9 The picture of produce chiller area (a), the boxing area (b) and the storage area (c) in supermarket C.

The main products that stored in this chiller room are shown in Table 2.6. Compared to other supermarkets surveyed, supermarket C usually only stored the products for a short period of time before putting them into the display area directly, so sometimes the chiller room was quite empty.

Table 2.6 Details of room and product inside for supermarket C.

Areas	Room size	Main products	Weight (kg)	Door
Chiller room	4.35 m × 3.56 m	Apple Mushroom Greens Grapes Citrus Blueberry	171 36 308 144 240 54	Without strip curtains
Boxing area	5.20 m × 1.80 m	Wheat flour	750	Without strip curtains
Storage area	12.00m × 3.00 m	Banana Tomato Potato	180 150 198	With strip curtains

2.2.5.5 Supermarket D

Considering the storage area is quite empty, only chiller room was chosen in supermarket D. The location of sampling area and I-button was shown in Figure 2.10. The sampling area was far away from the loading zone and not any petrol exhausted machine was used in supermarket D.

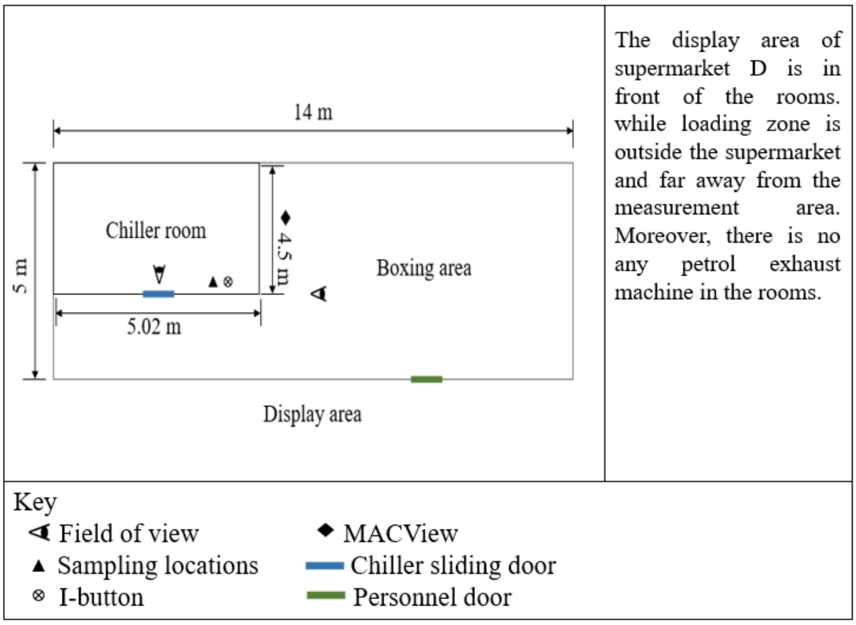


Figure 2.10 Plan of supermarket D.

Figure 2.11 The picture of produce chiller area (a) and the storage area (b) in supermarket D.

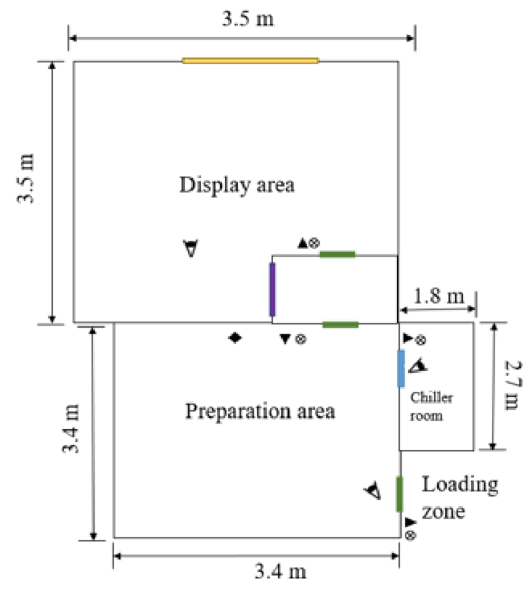
All the products were stored in the chiller room for a very short time and then they were moved to display area directly (Table 2.7). Moreover, 144kg apples were stored in the chiller room just for three days during the trial.

Table 2.7 Details of room and product inside for supermarket D.

Area	**Room size**	**Main products**	**Weight (kg)**	**Door**
Chiller room	4.50 m × 5.02 m × 2.93 m	Apple Peach Plum Pear Grape Tomato Strawberry Blueberry Citrus Carrot Greens	144 48 32 144 30 30 30 36 25 330	Without strip curtains

2.2.5.6 Flower distributor

The layout of the flower distributor was shown in Figure 2.12. and Figure 2.13., and 4 locations are chosen to be tested, which includes the preparation area, display area, chiller room and ambient. There was not petrol exhausted lift in the flower distributor.



The flower distributor is located in highway area. Loading zone is on the right side of the preparation area, and it is in isolation. In addition, only privacy cars park in the loading zone and there is no lifts in the flower distributor.

Key
- Field of view
- Sampling locations
- I-button
- MACView
- Chiller sliding door
- Main gate
- Personnel door
- Window

Figure 2.12 Plan of the flower distributor.

Figure 2.13 The picture of produce chiller area (a) and the preparation area with room temperature (b) and the display area (c) in flower distributor.

The main products in the chiller room were roses, cordyline and daffodils (Table 2.8). The products off the preparation area are mainly green plants and stock, which were not a lot. Sometimes the flowers would be stored this area, as the new products were always transferred from the gate in the preparation area, and it took a while before the flowers were placed in the chiller room and display area. All of these areas did not have any plastic curtain and the main gate of the flower distributor, which was connected with the display area directly, were usually open during working hours (9am – 5pm??). Furthermore, gerbera, roses and lilies are the main products of the display area.

Table 2.8 Details of room and product inside for flower distributor.

Areas	Room size	Main products	Branches	Door
Chiller room	2.70 m × 1.80 m × 2.00 m	Roses Carnation Cordyline Daffodils Ranunculus Leptospermum Lilies	20 8 12 10 5 6 8	Without strip curtains
Preparation area	3.40 m × 3.40 m × 2.30 m	Green plants Stock	7 4	Without strip curtains
Display area	3.55 m × 3.55 m × 2.30 m	Roses Daffodils Lilies Dracaena Ranunculus Tulip Protea Alstroemeria Gerbera	35 10 28 10 11 7 7 14 47	Without strip curtains

2.3 Results

2.3.1 Distribution Centre

The results show that the average temperature of all the chiller rooms were around 4.5°C in the distribution centre (Figure 2.14). It is expected that the range of ethylene concentration in the ambient area was the lowest, which were from 0 nL L^{-1} to 136 nL L^{-1}. Although the peak value of ambient was 136 nL L^{-1}, the median and 80^{th} percentile of the ambient area were 10 nL L^{-1} and 20 nL L^{-1} respectively. As the sampling area was set near the loading zone and the ambient area was not be isolated, the air pollution, such as smoking and petrol exhausting, can cause an increase of ethylene. Chiller rooms 2 (0 – 1353.27 nL L^{-1}) and 3 (0 – 1343 nL L^{-1}) had similar ranges of ethylene concentration. The median and 80th percentile in chiller room 2, which were 93 nL L^{-1} and 126 nL L^{-1} respectively, were slightly higher than those in chiller room 3 (median: 86 nL L^{-1}, 80th percentile: 116 nL L^{-1}). The main products in these rooms were green leafy and root crops which did not emit a lot of ethylene in these two rooms. Chiller room 4 had the lowest peak measured of ethylene (477.23 nL L^{-1}). Only a small amount of mushroom and red onions, which only produce a few ethylene concentrations, stored there could be the reason why the ethylene concentration was lower than other chiller rooms. Besides, the median and 80th percentile of ethylene were 102 nL L^{-1} and 137 nL L^{-1}. The ranges of ethylene concentration in chiller rooms 5 and 6 were much wider, being 0 – 3393 nL L^{-1} and 0 - 3678.4 nL L^{-1} respectively. For chiller room 5, the median and 80th percentile were 225 nL $L^{-1}$and 752 nL L^{-1}, lower than the 374 nL L^{-1} and 964.5 nL L^{-1}found in chiller room 6. The chiller room 6 was filled with huge amounts of apples, while some peaches and avocados were stored in chiller room 5. Apples, avocados and peaches are high ethylene producing products, so the ethylene concentration in these two rooms was quite high.

The ethylene concentration fluctuated throughout the day (Figure 2.15a – b; Figure A.1a-1d). The ethylene concentration from 9 am to 9 pm on Saturdays and Sundays increased significantly, it is a result of all the doors being closed, limiting gas exchange. Additionally, ethylene of all the locations increased from 8 pm to 5 am during weekdays. The doors being closed at nights and reopen the next morning can account for this phenomenon. However, there was no ethylene accumulation observed in room 3 during the weekends and working days. It is possible that not a lot of products were stored in this room, so the ethylene concentration remained relatively stable. In the graphs (Figure 2.15a – b; Figure A.1a-1d), there were some single peaks occurred. This could be due to a sudden surge of gas exhaust while operating the petrol forklift near the storage room. Generally speaking, the trend of ethylene was quite stable from 8 pm to 5 am. The ethylene concentration in the ambient area also followed this trend, as it was placed nearby the door (Figure 2.15b). Some trunks and petrol forklifts parked nearby during closing time, so the ethylene concentration can be built up. The storage room temperature also fluctuated during the day. When the products were transported for distribution, the door of the chiller room was open, causing heat exchange with storage area 1 (Fig. 2.2) the which had a higher average temperature.

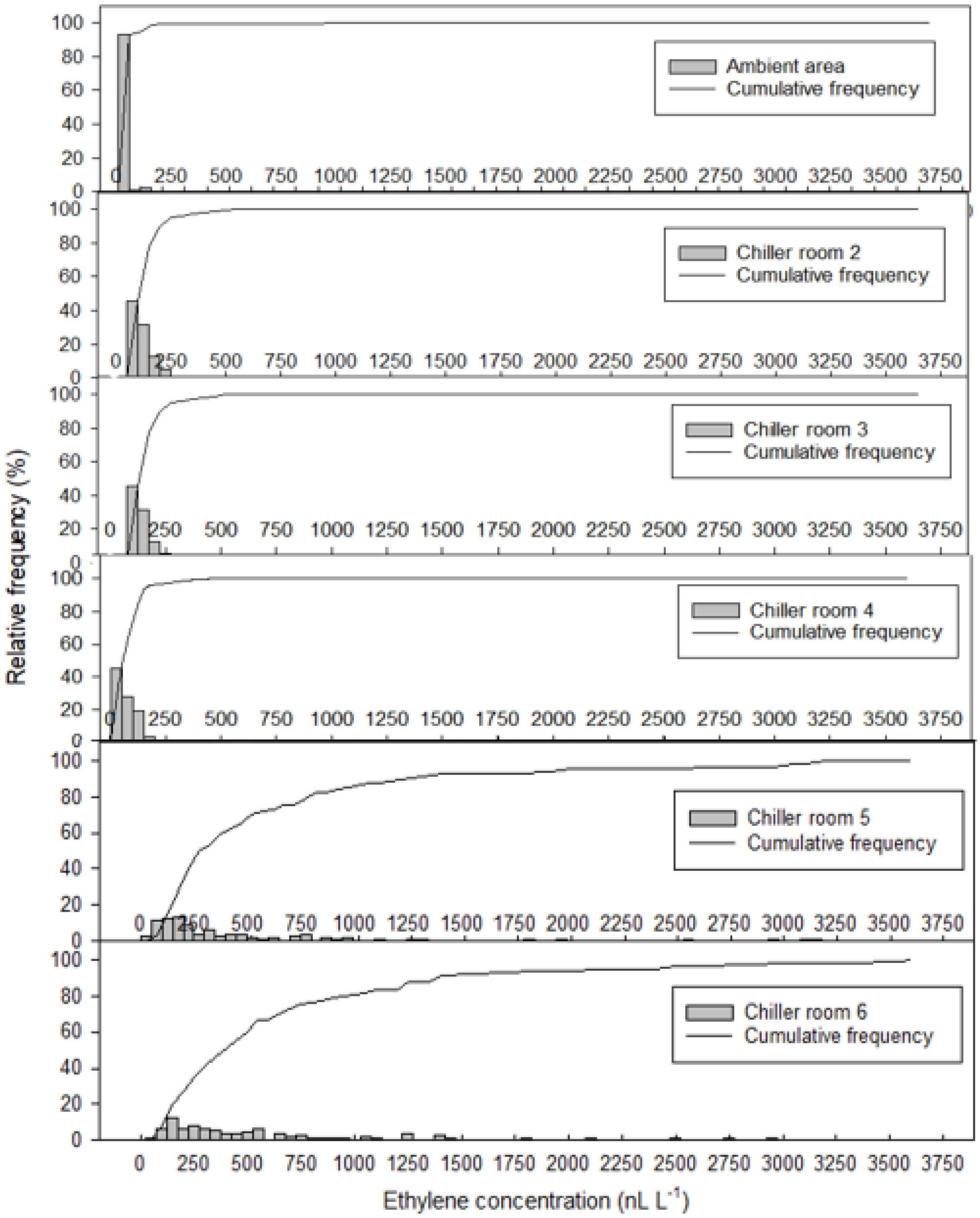


Figure 2.14 Histogram bar and cumulative frequency line of ethylene concentrations measured in different rooms at the distribution centre. Locations are (a) ambient area; (b) chiller room 2; (c) chiller room 3; (d) chiller room 4; (e) chiller room 5 and (f) chiller room 6 as shown in Figure 2.2. Each location contains 3 data points over a 45 min/h period (15min per reading), entire data last 4 weeks from 13th May to 10th June.

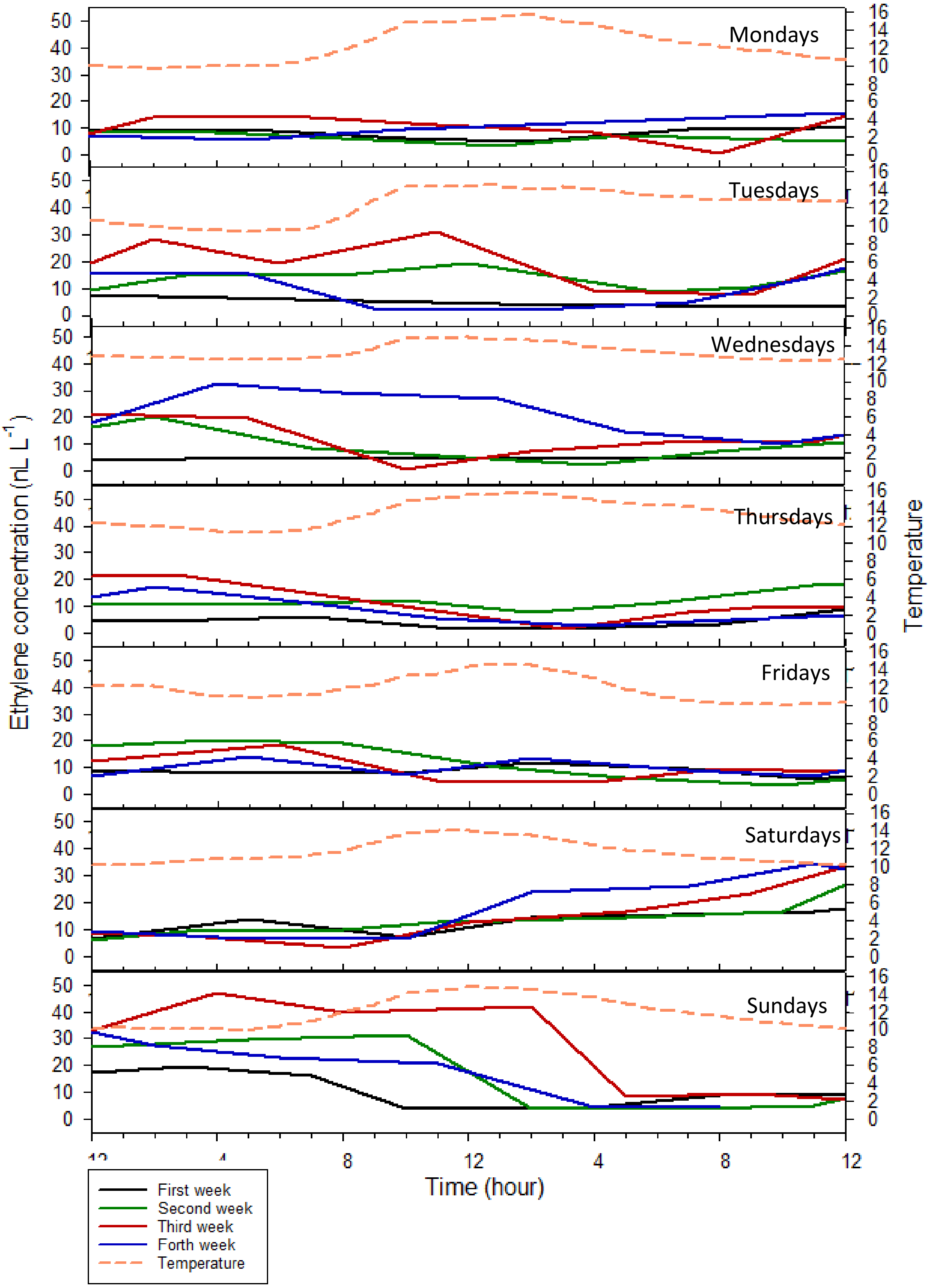


Figure 2.15a The daily trend of average ethylene concentration and the average temperature of ambient area in distribution centre. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 4 weeks from 13th May to 10th June.

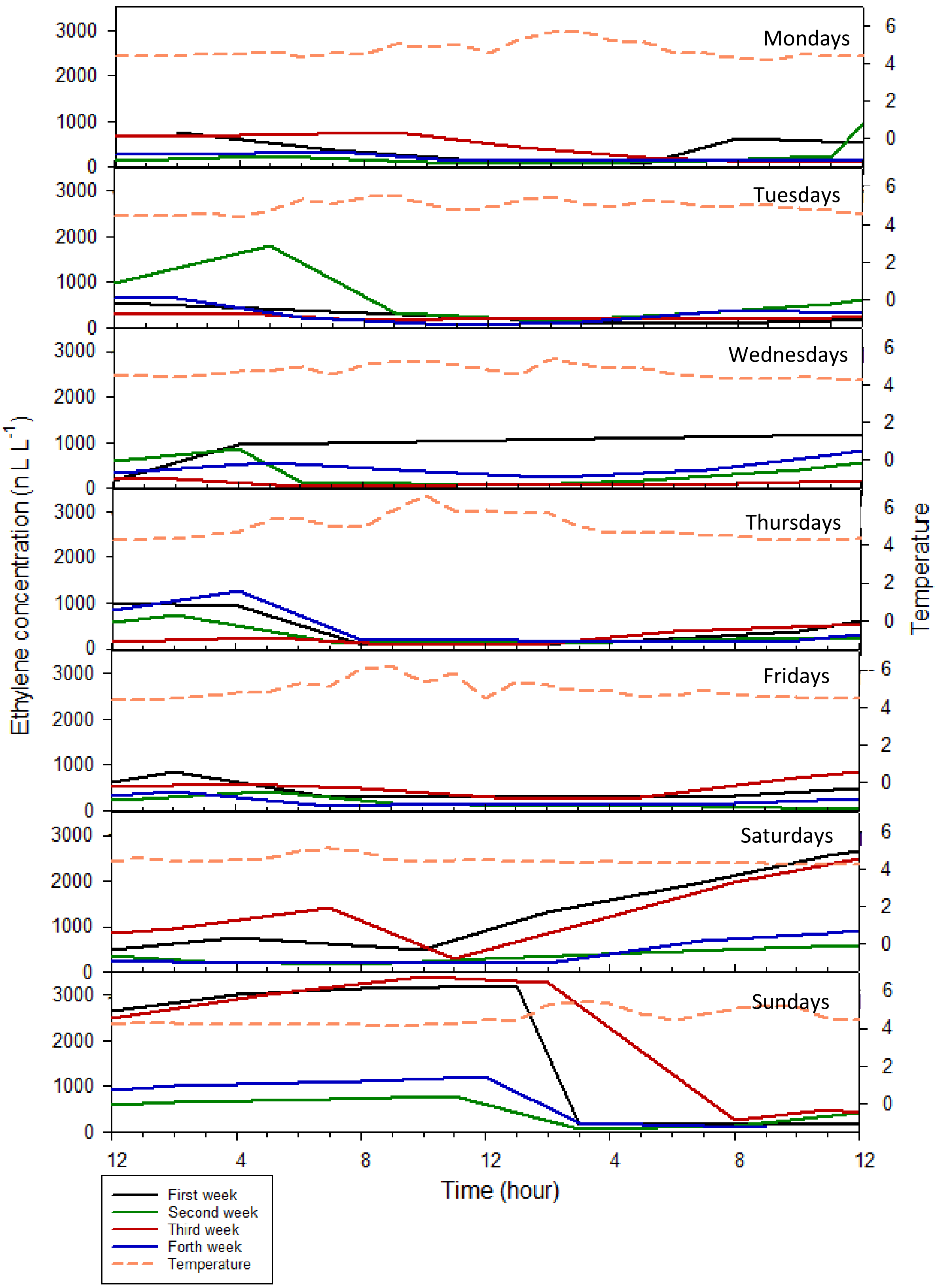


Figure 2.15b The daily trend of average ethylene concentration and the average temperature of chiller room 5 (citrus) in distribution centre. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 4 weeks from 13th May to 10th June.

2.3.2 Supermarket A

The ambient temperature averaged at 15°C. Not unexpectedly the ambient area had the lowest ethylene concentrations, ranging from 0.25 – 60.5 nL L^{-1} (Figure 2.16). The chiller room and display area operated on average at 4 °C and 12.3 °C respectively. The chiller room and display areas were found to have very similar ethylene concentration profiles (Figure 17b-c). Ethylene concentration in the chiller room ranged from 3.45 – 182.33 nL L^{-1}, while the storage area ranged from 10.37 – 208.67 nL L^{-1}. The weight of high ethylene production products in the chiller room and display area was similar (190kg apples in the chiller room and 88kg avocados and 80kg pears in storage area. For both rooms, ethylene concentrations were skewed towards the lower end of the range. The median and 80th percentile for the chiller room and storage area were 29 and 84 nL L^{-1}, and 33 and 51 nL L^{-1} respectively.

Generally, the ethylene concentration in the ambient area (inside the room) (Figure 2.17a), chiller room (Figure 2.17b) and storage area (Figure A.2) were higher from 6 pm to 5 am, as the closed door impacted air ventilation. At 5 am, the door was open for the start of the working day and hence ethylene started to decline from this time until 8 am. The ethylene concentration in the ambient area remained almost exclusively < 10 nL L^{-1} between 8 am and 8 pm daily (Figure 2.17a). The ethylene concentration frequently accumulated during the night from 8 pm peaking at approximately midnight, consistently occurring on a Wednesday. As the ambient area was located inside the room, closed door at 6pm could also impact the air ventilation and the ethylene built-up during the night. The ethylene concentration in the chiller room from 8 am to 8 pm fluctuated in comparison to other locations. The fluctuation occurred because the delivery of products was more frequent in the chiller room during this time. The ethylene level of storage area had a single peak value at 208.67 nL L^{-1}, much higher than the value at 80th percentile (51 nL L^{-1}). A possible reason for this isolated measurement is that on occasions a hydraulic lift passed the area and hence could cause an increase of ethylene concentration.

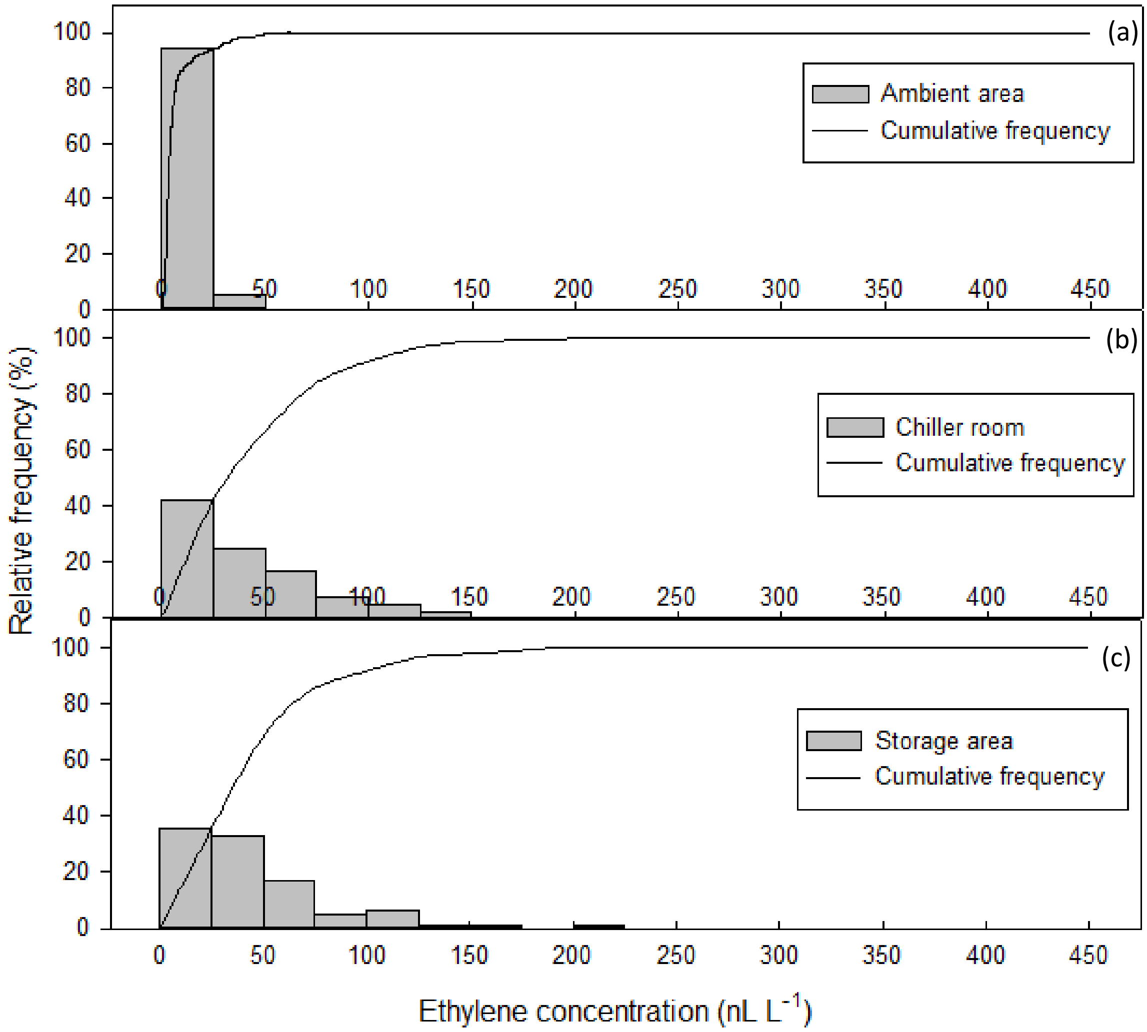


Figure 2.16 Histogram bar and cumulative frequency line of ethylene concentrations measured in different rooms at the supermarket A. Locations are (a) ambient area; (b) chiller room and (c) storage area as shown in Figure 2.4. Each location contains 3 data points over a 45 min/h period (15min per reading), entire data last 3 weeks from 17th July to 6th August.

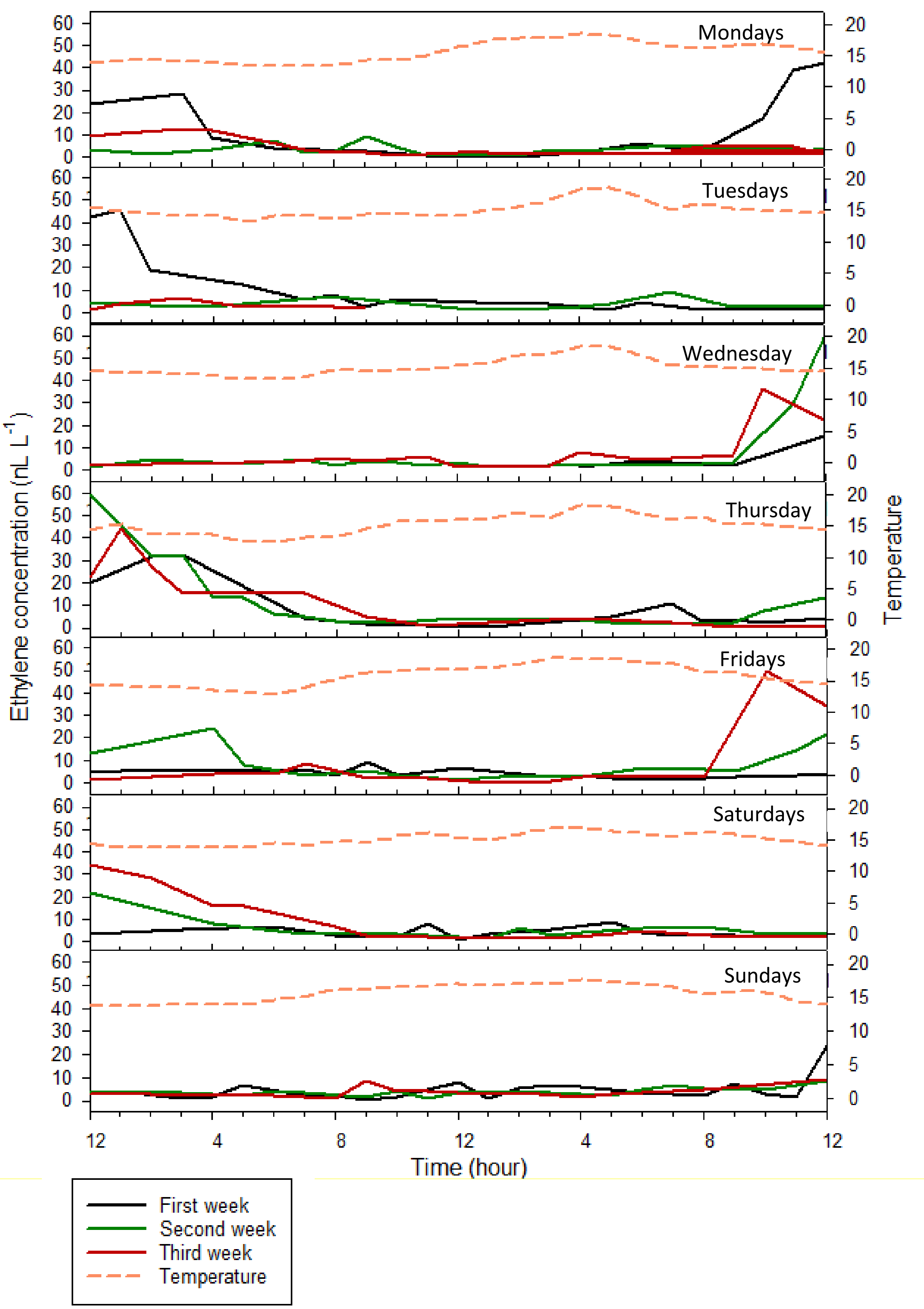


Figure 2.17a The daily trend of average ethylene concentration of ambient area in supermarket A. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 3 weeks from17th July to 6th August.

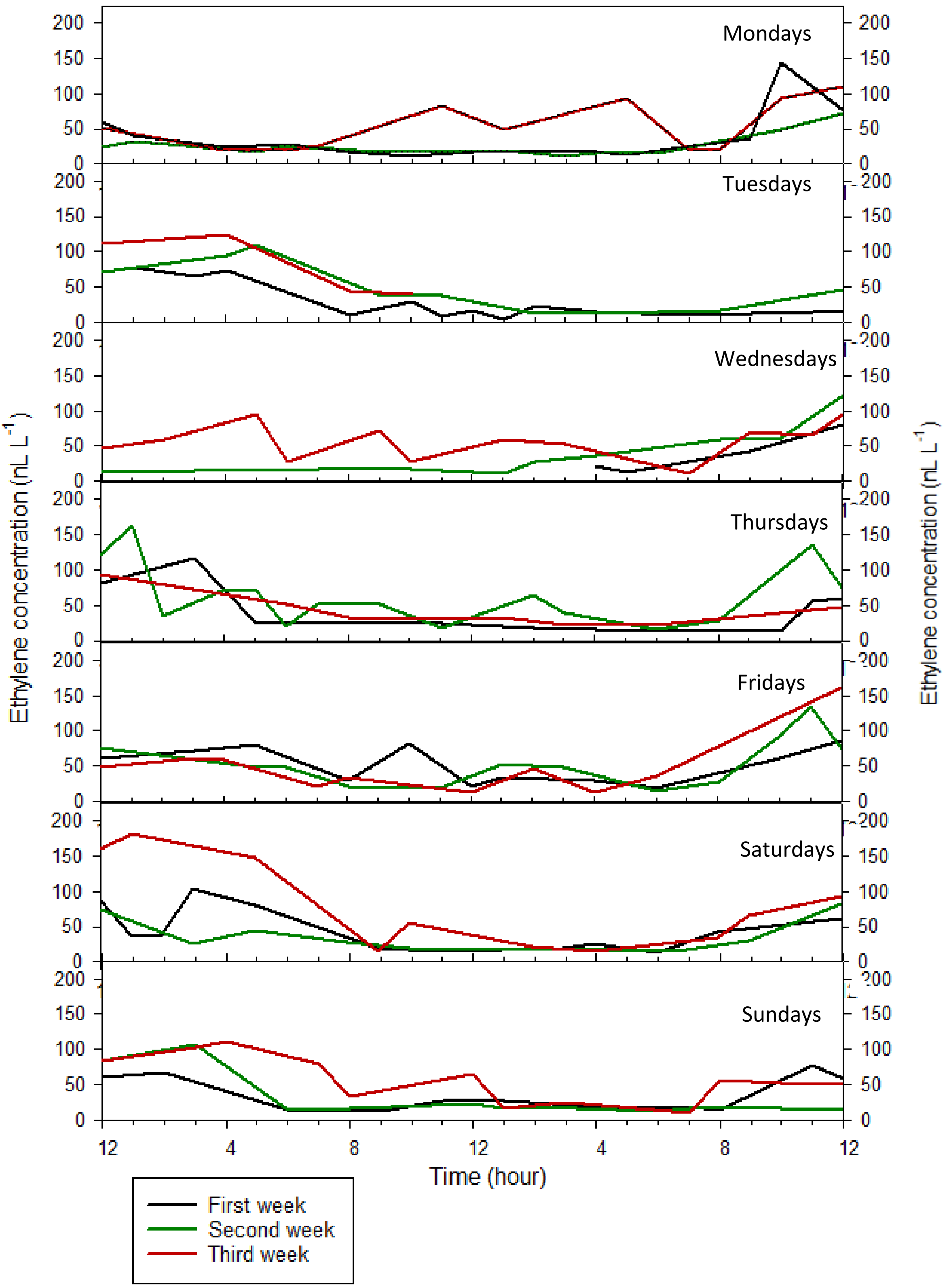


Figure 2.17b The daily trend of average ethylene concentration of chiller room in supermarket A. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 3 weeks from 17th July to 6th August.

2.3.3 Supermarket B

The ambient area, chiller room, banana area and shelf area were on average at 12.28°C 4.16 °C, 13.5 °C and 12.52 °C respectively. The ethylene level in the ambient area of supermarket B averaged at below 50nL L^{-1} (around 83%) and it ranged from 0 to 100nL L^{-1}.

The ethylene concentration in the ambient area ranged between 0.33 and 97.27 nL L^{-1} (Figure 2.18), with 80% of the ethylene concentration detected at around 22 nL L^{-1}. The range of ethylene concentration in the chiller room was from 20.3 nL L^{-1} to 464.1 nL L^{-1}, with the medium and 80^{th} percentile being 81 and 207 nL L^{-1} respectively. The ethylene concentration in the chiller room had a wider range compared to other areas, as some high ethylene production fruits were stored here, such as citrus and apples. The banana area and the shelf area had similar ethylene concentrations, with the banana area having a slightly wider range (2.6– 107.27 nL L^{-1}) than that in the shelf area (4.27–85.6 nL L^{-1}). Both had the median and 80^{th} percentile of 29.5 and approx. 48 nL L^{-1} respectively. These two areas shared the same space and the banana was the main products to produce ethylene in this area. The banana area was near to the loading area, and the combustion products were nearby which could also have increased the measured ethylene concentration.

The ethylene concentration from all the locations increased slightly after 6pm until the next early mornings around 5 am (Figure 2.19a – b; Figure A.3). The door closed during this period of time and it can build up the ethylene. From 8 am to 6 pm, the ethylene concentration from all the locations were quite stable. However, the ethylene concentration in shelf area on Sundays fluctuated possibly because the transport of products was more frequently on Sundays in preparation for working on Monday.

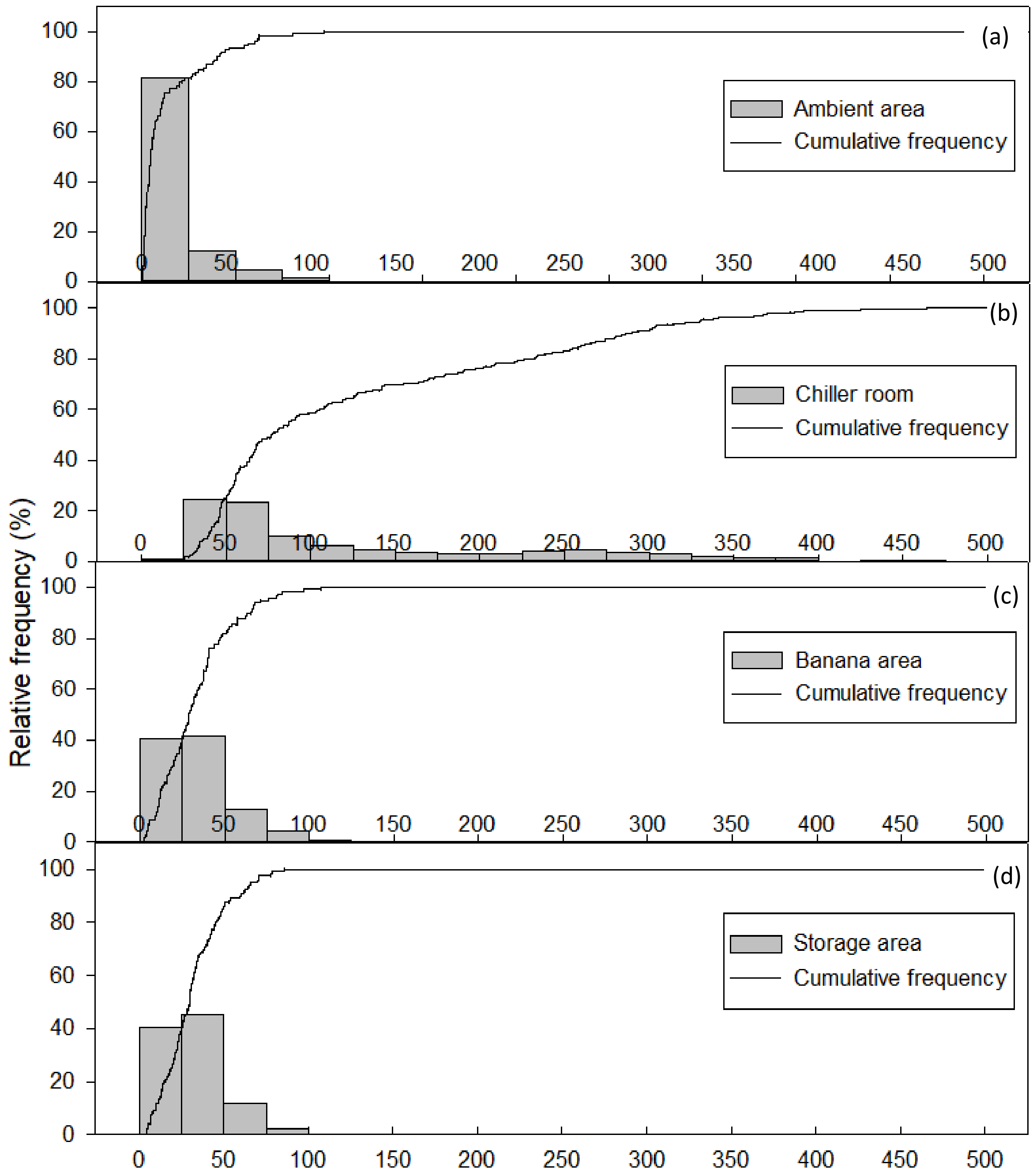


Figure 2.18 Histogram bar and cumulative frequency line of ethylene concentrations measured in different rooms at the supermarket B. Locations are (a) ambient area; (b) chiller room; (c) banana area and (d) storage area as shown in Figure 2.6. Each location contains 3 data points over a 45 min/h period (15min per reading), entire data last 3 weeks from 7th August to 4th September.

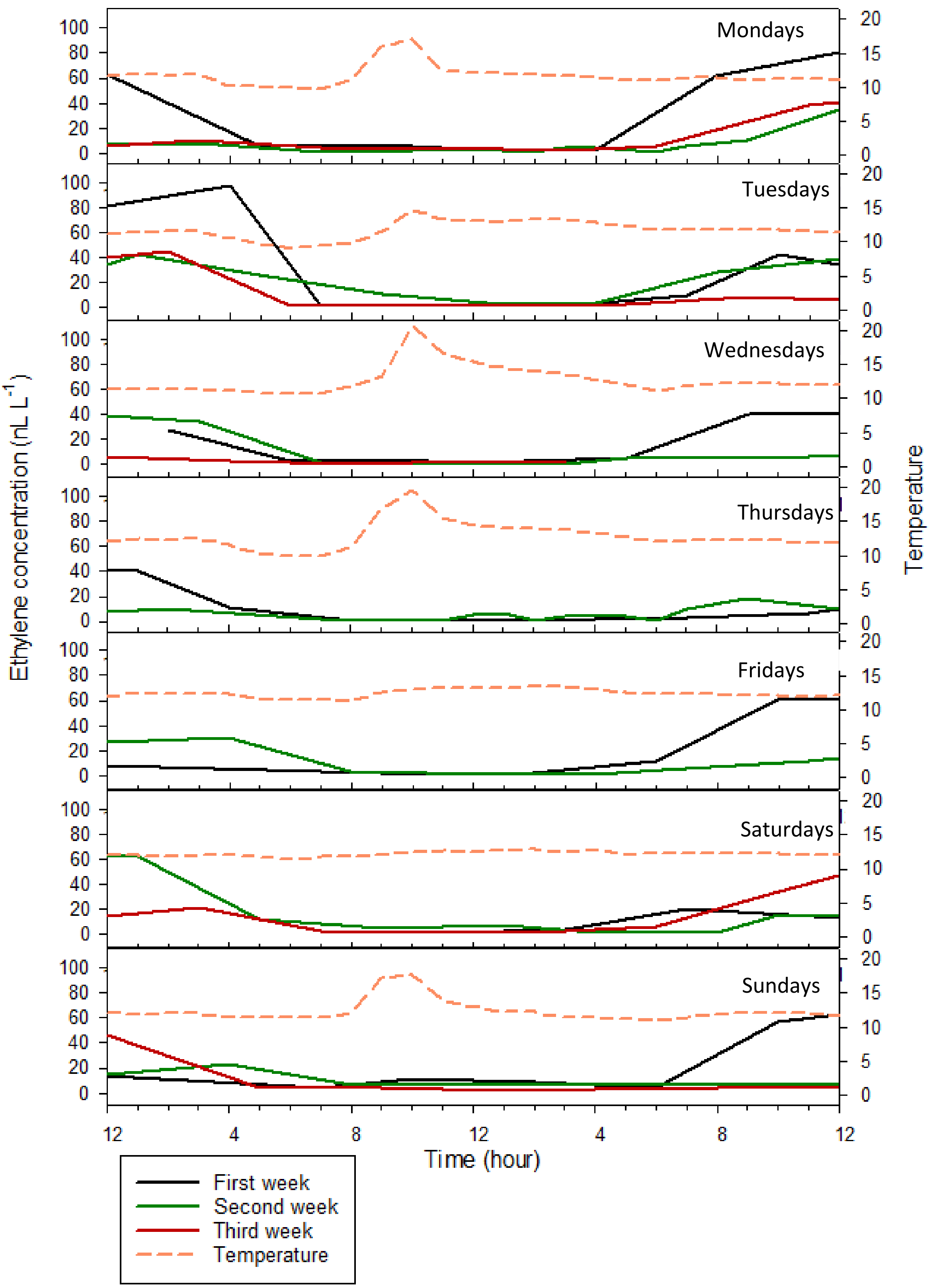


Figure 2.19a The daily trend of average ethylene concentration and the average temperature of ambient area in supermarket B. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 3 weeks from 7th August to 4th September.

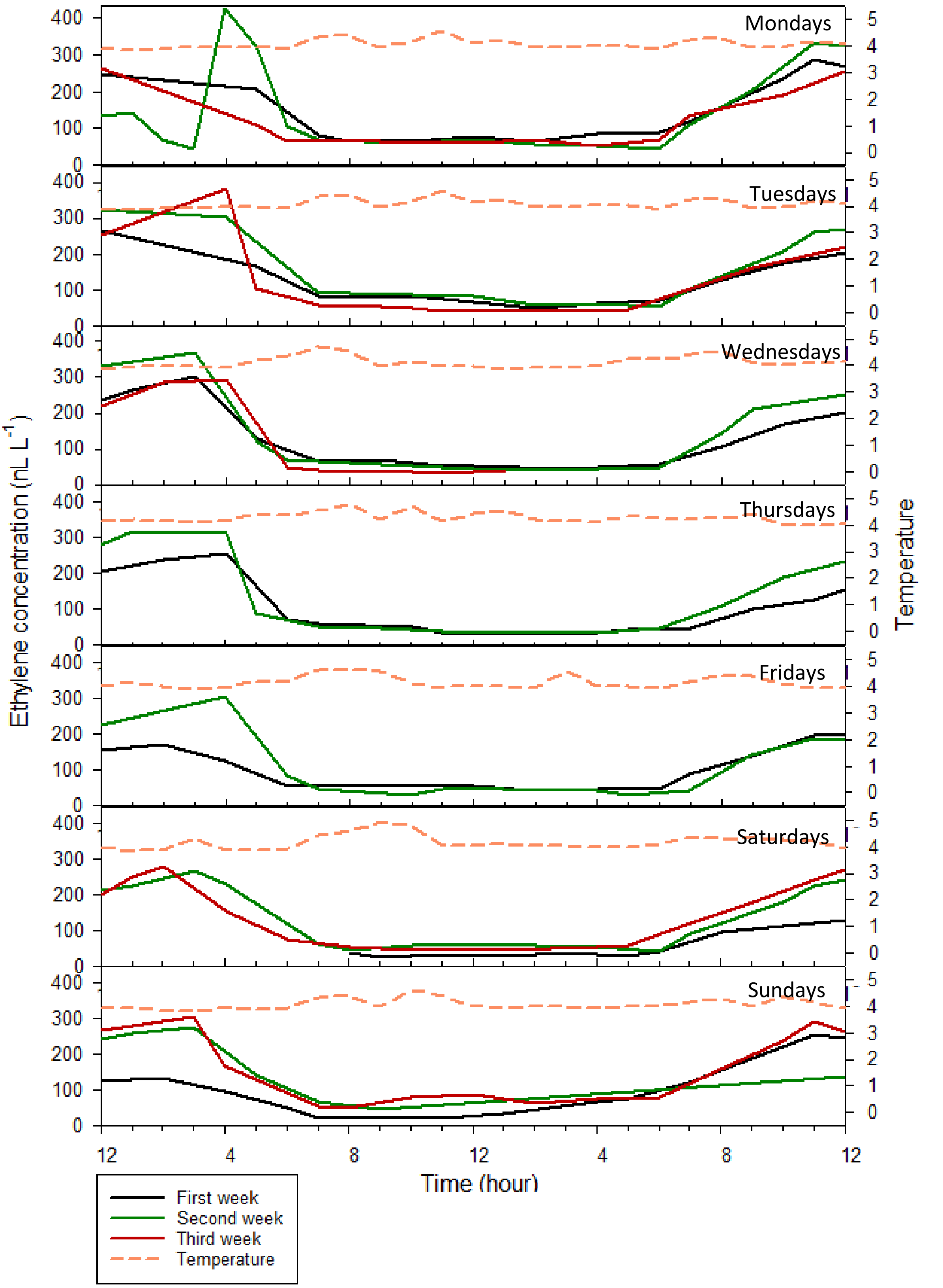


Figure 2.19b The daily trend of average ethylene concentration and the average temperature of chiller room in supermarket B. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 3 weeks from 7th August to 4th September.

2.3.4 Supermarket C

For supermarket C, the ethylene concentration in chiller room, boxing area and storage area had similar ranges (0 – approx. 400 nL L^{-1}) (Figure 2.20). For chiller room (with average temperature 4.23 ℃), the median and 80^{th} percentile were 6.5 and 27 nL L^{-1}. Also, the median and 80^{th} percentile were 6 and 16 nL L^{-1} in the boxing room (with average temperature 16.47 ℃), while they in the storage area (with average temperature 16.47 ℃) were 2.5 and 13 nL L^{-1}. The boxing area and storage area shared the same space and hence the ethylene concentration was similar. Supermarket C only stored products in the chiller room for a short time and then the products were put in the display area directly. This can be the reason why the ethylene concentration in these three locations were similar. However, the highest value of ethylene occurred in the storage area, which was 393 nL L^{-1}. More liftings were stopped nearby that can account for this result. As many studies reported that air pollution, such as fossil fuels, smoking, burning, can cause an increase of ethylene.

The ethylene concentration always peaked at 6 am in the morning, considering the hydraulic lifts were used to deliver the products and parked nearby during those hours (Figure 2.21a – b; Figure A.4). Moreover, the ethylene concentration in the boxing and storage area had a higher fluctuation than chiller room, possibly because these two locations were in close proximity to the loading zone.

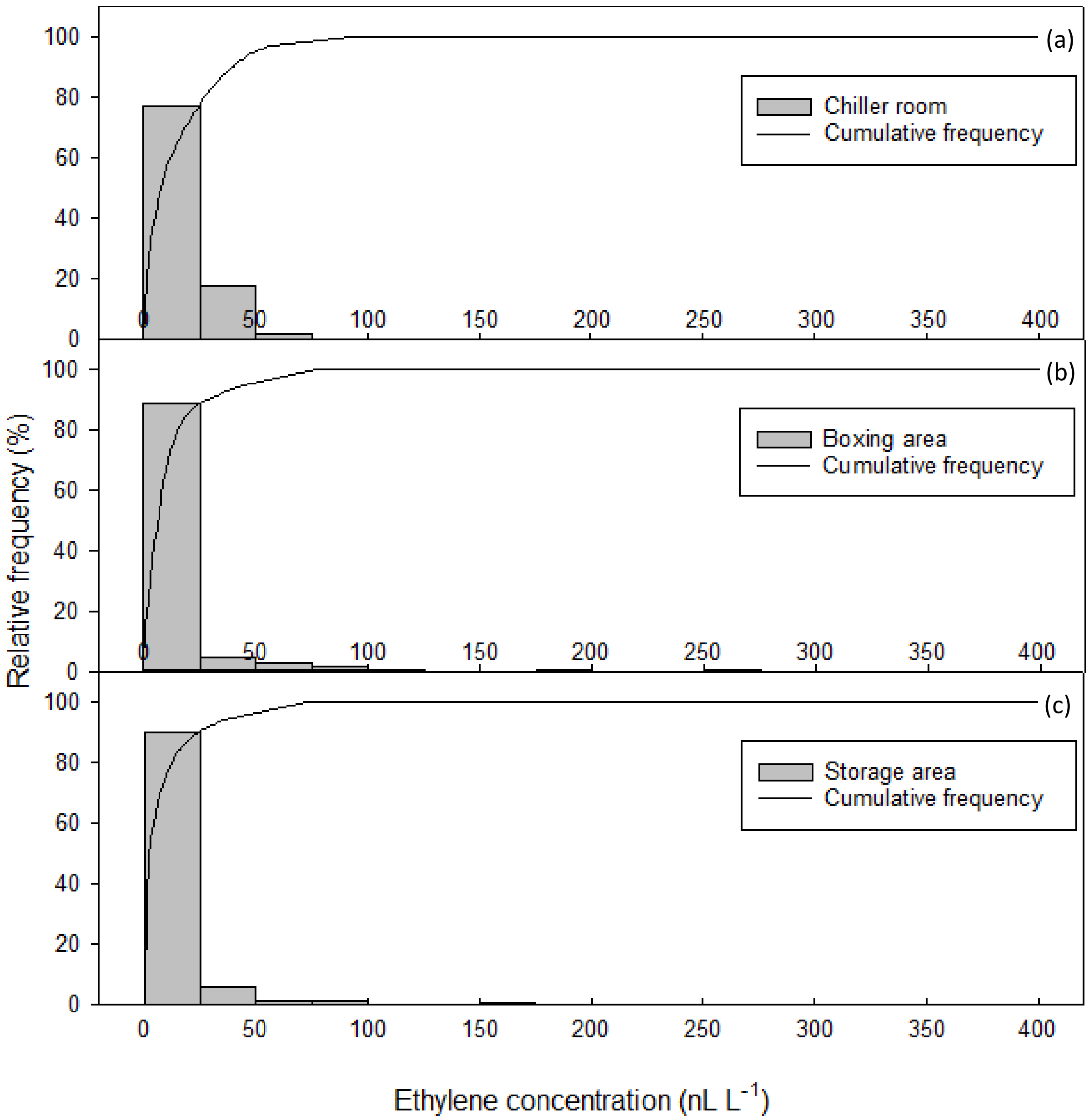


Figure 2.20 Histogram bar and cumulative frequency line of ethylene concentrations measured in different rooms at the supermarket C. Locations are (a) chiller room; (b) boxing area and (c) storage area as shown in Figure 2.8. Each location contains 3 data points over a 45 min/h period (15min per reading), entire data last 5 weeks from 8th November to 4rd December. From 21th November to 27th November, only the ethylene concentration in chiller room was measured.

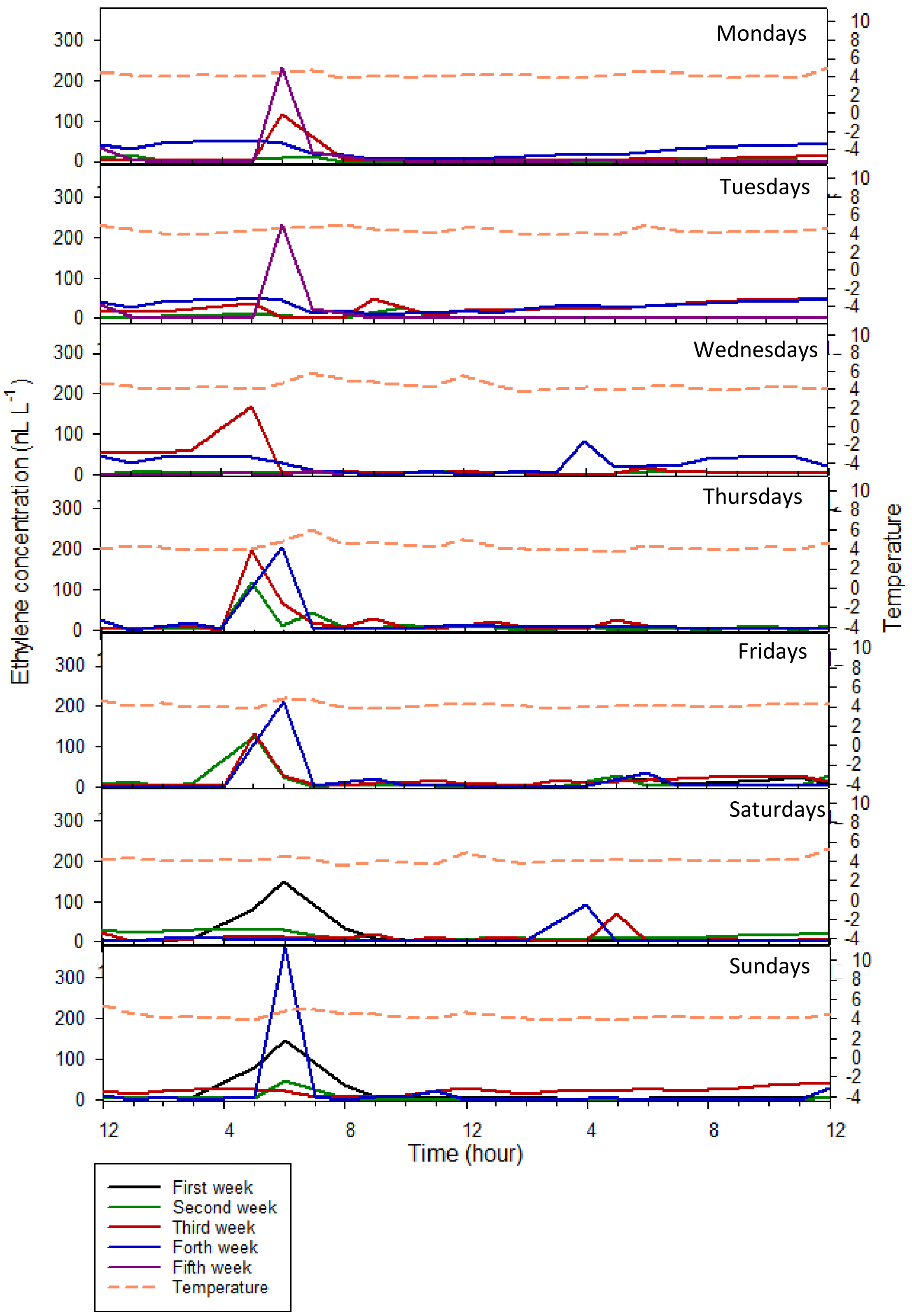


Figure 2.21a The daily trend of average ethylene concentration and the average temperature of chiller room in supermarket C. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 5 weeks from 8th November to 4rd December.

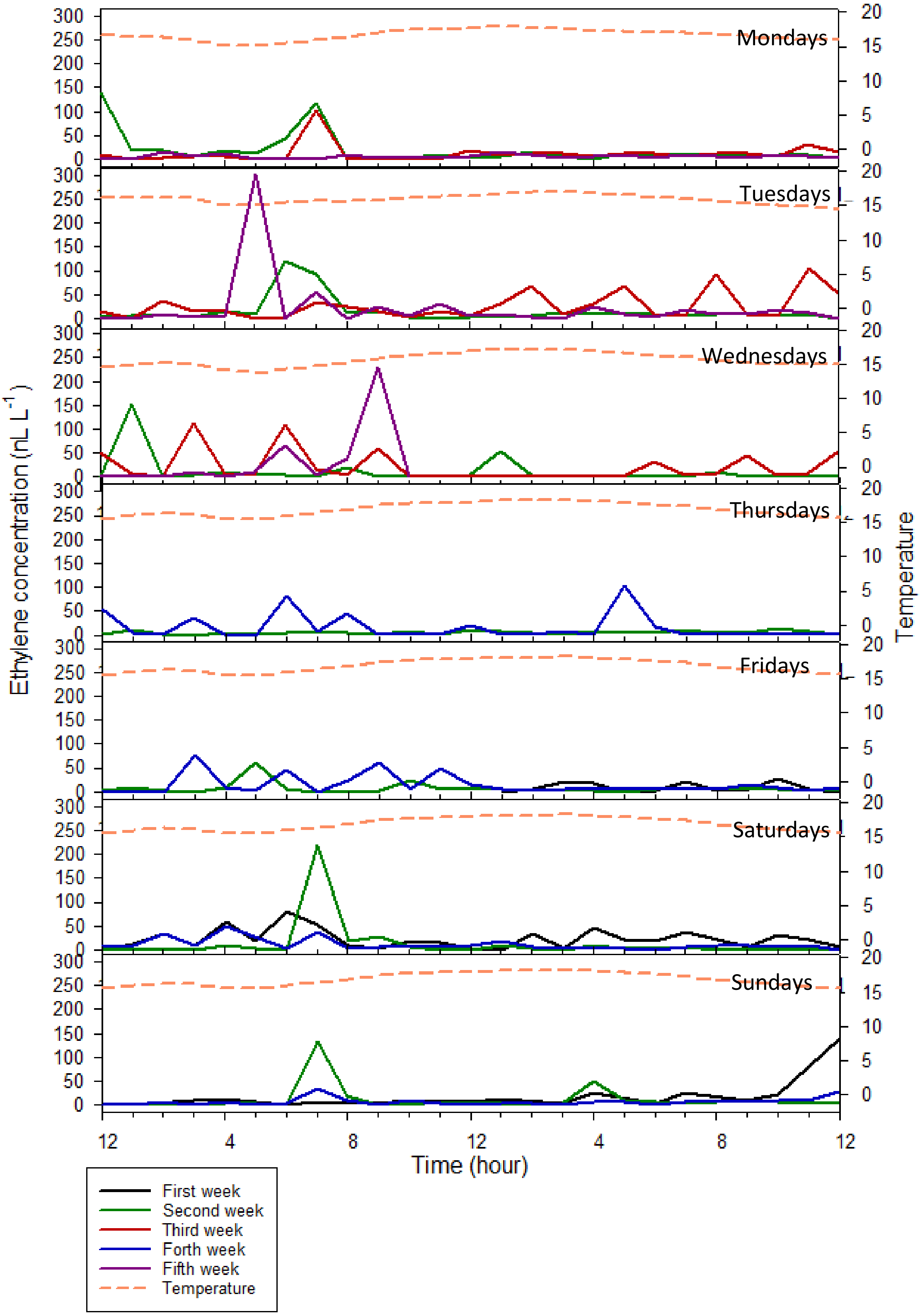


Figure 2.21b The daily trend of average ethylene concentration and the average temperature of boxing area in supermarket C. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 5 weeks from8th November to 4rd December.

2.3.5 Supermarket D

The average temperature in the chiller room was 4.11℃. The ethylene concentration ranged from 1.4 nL L^{-1} to 115.4 nL L^{-1} (Figure 2.22), with the median and 80% percentile being 16.5 and 38 nL L^{-1}, respectively. There were no sharp peaks of ethylene concentration observed in supermarket D as the supermarket was far away from the loading area, and hence less likely to have air pollution due to smoking and the using of hydraulic lifts and other fuel oil or coal combustion.

The ethylene concentration climbed slightly between 6pm and 5am while the supermarket was closed, and the doors were kept shut (Figure 2.23). The doors were open at 5 am and the products were usually delivered to the chiller room from 8 am, and then delivered to the display area at around 12 pm. Therefore, the ethylene concentration increased from 8 am and dropped down again from 12 pm. The ethylene concentration on Sunday of forth week and Monday of the fifth week were below 40 nL L^{-1}, possibly because there were not many ethylene production products stored here.

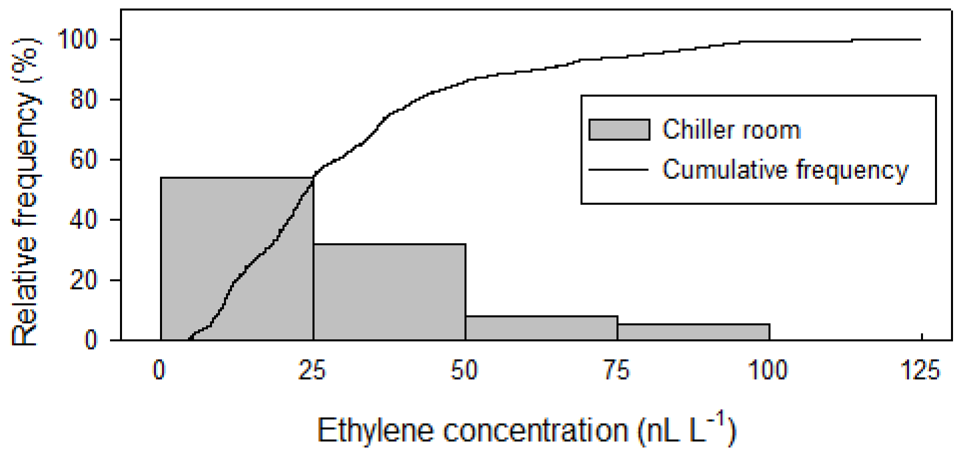


Figure 2.22 Histogram bar and cumulative frequency line of ethylene concentrations measured in chiller room at the supermarket D. Location is shown in Figure 2.10. Each location contains 3 data points over a 45 min/h period (15min per reading), entire data last 5 weeks from 9th January to 7th February.

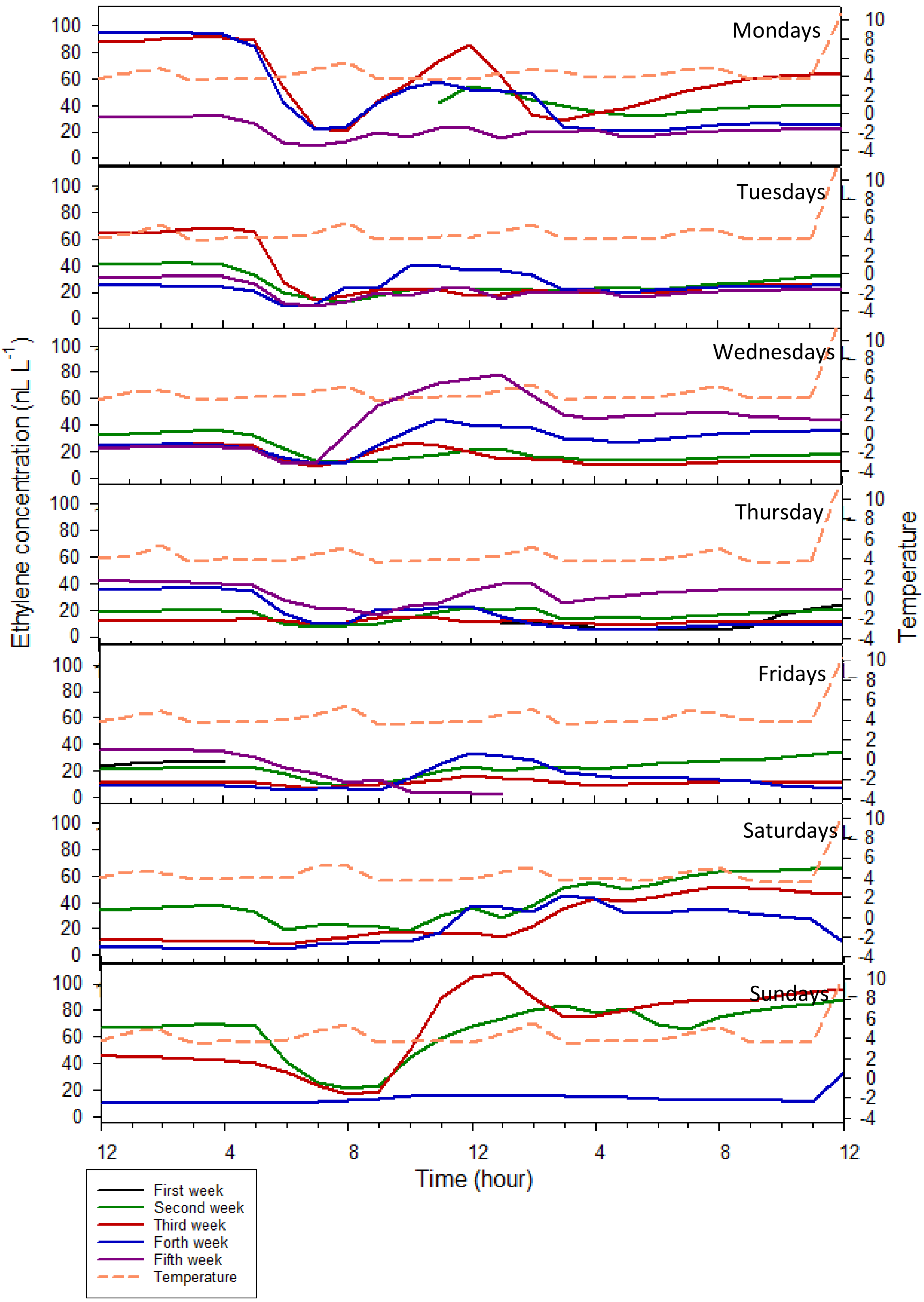


Figure 2.23 The daily trend of average ethylene concentration and the average temperature of chiller room in supermarket D. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 5 weeks from 9th January to 7th February.

2.3.6 Flower distributor

In the experiment, the ethylene concentration of different areas in the flower distributor were all below 10nL L^{-1}, which were similar to the ambient area (Figure 2.24). The ethylene concentration range in ambient (with average temperature 10.89℃), chiller room (with average temperature 4.22℃), preparation area (with average temperature 15.19℃) and display area (with average temperature 14.23℃)were 0.03 - 6.53 nL L^{-1}, 0.1 - 9.73 nL L^{-1}, 0.17 - 6.73 nL L^{-1} and 0.2 - 6.17 nL L^{-1} respectively. Furthermore, the 80th percentile for all the locations was below 5 nL L^{-1}.

The ethylene increased from 6 pm to midnight from Mondays to Fridays, due to the closed door reduced the gas ventilation (Figure 2.25). The ethylene concentration was quite stable in all the locations from 5 am on Saturdays till the end of Sundays. This phenomenon can be accounted for that there were no products delivering during the weekends. However, most products were moved around and delivered between 7 am and 6 pm, hence the ethylene concentration was more fluctuated during the period time. Some peaks were showed in Figure 2.25 and Figure A.5a - c. It was caused by unknown reason. Two possible reasons could account for this. Firstly, the flower distributor is located to the main street, traffic and car park for a short time can be caused the changes in ethylene. Another possible reason was that the ethylene stickes can be a reason, considering the first reading of MACView can be impacted by the last reading from the previous room. As the highest value was 9.73 nL L^{-1}, it was still extremely low and cannot cause a risk.

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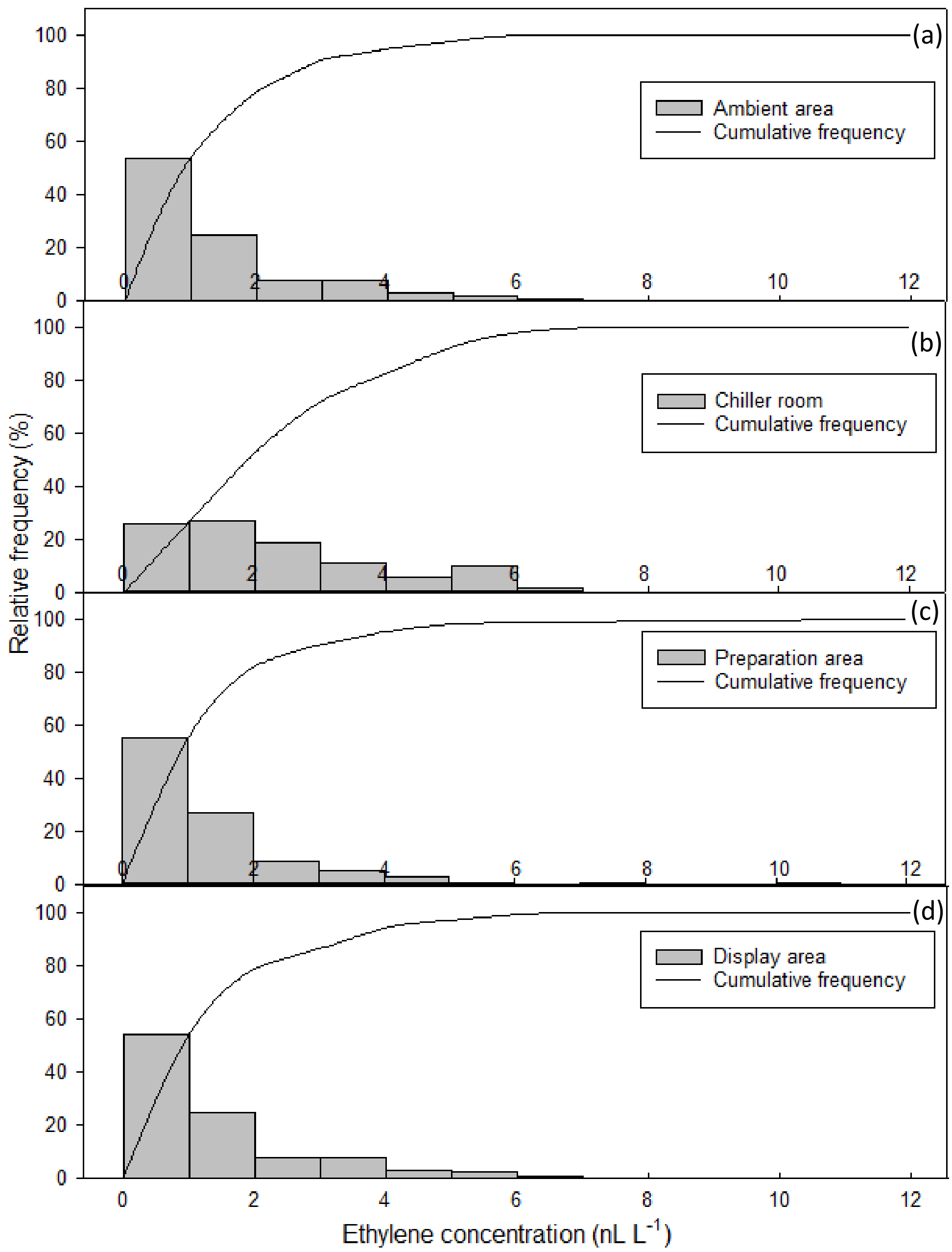


Figure 2.24 Histogram bar and cumulative frequency line of ethylene concentrations measured in different rooms in flower distributor. Locations are (a) ambient area; (b) chiller room; (c) preparation area and (d) display area as shown in Figure 2.12. . Location is shown in Figure 2.10. Each location contains 3 data points over a 45 min/h period (15min per reading), entire data last 4 weeks from 3rd September to 28th September.

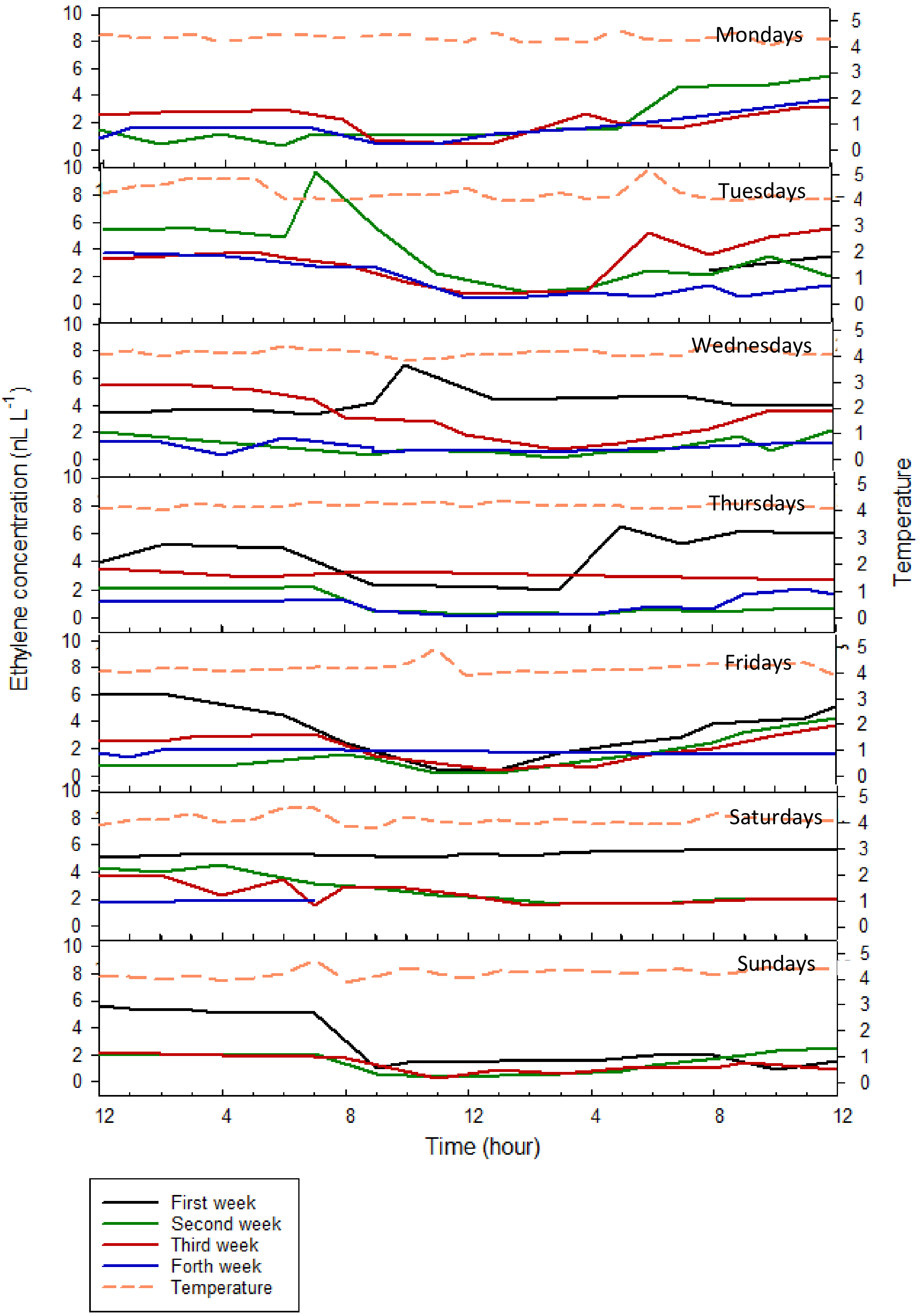


Figure 2.25 The daily trend of ethylene concentration and the average temperature of chiller room in flower distributor. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 4 weeks from 3rd September to 28th September.

2.4 Overall Discussion

2.4.1 Ambient Ethylene Concentrations

Table 2.9 Overview of ethylene concentration survey results.

Site	Locations measured	Location	Peak Measured (nL L^{-1})	Median (nL L^{-1})	80^{th} Percentile (nL L^{-1})
Distribution Centre	6	Ambient	136.2	12.55	20
		2	1353.27	105.22	116
		3	1343	87.35	126
		4	477.23	112.42	137
		5	3393	590.9	752
		6	3678.4	712.35	964.5
Supermarket A	3	Ambient	60.5	18.31	7
		Chiller room	182.3	47.21	84
		Storage area	208.7	47.65	51
Supermarket B	4	Ambient	92.3	13.18	22
		Chiller room	464.1	125.9	207
		Banana area	107.3	32.59	48
		Shelf area	85.6	31.52	46
Supermarket C	3	Chiller room	375.7	17.04	27
		Boxing area	361.4	15.17	16
		Storage area	318.6	11.95	13
Supermarket D	1	Chiller room	115.4	29.79	38
Flower Distributor	4	Ambient	6.53	0.88	1.3
		Chiller room	9.73	2.34	3.8
		Preparation	6.73	1.35	2.2
		Display area	6.17	1.4	2.2

The 80^{th} percentile ethylene concentrations in ambient areas of distribution centre, supermarkets and flower distributor ranged from 1.3 – 22 nL L^{-1} (Table 2.9). This result is lower than that reported by the Wills et al. (2000) where ambient ethylene in three distribution centres ranged from 31 to 72 nL L^{-1}. However, the peak measurement of ethylene in supermarkets can reach more than 60 nL L^{-1} in supermarkets, while peak measured in distribution centre can reach 136.2 nL L^{-1}. The peak measured was found in the early morning. Ethylene built up during the closing time due to the limitation of gas exchange could be a reason (Wheeler et al., 2004). Moreover, most trunks and lifts parked nearby could be another reason. This is likely due to anthropogenic sources can produce ethylene (Sawada et al., 1989).

Abeles and Heggestad (1973) found that the ethylene in the ambient environment can range from 39 nL L^{-1} to 700 nL L^{-1} due to the location and air pollution. Luria et al. (2000) also found that the ethylene in ambient area with flue gas was more than 1500 nL L^{-1}. Keller et al. (2013) found that ethylene concentration in the air usually was below 5 nL L^{-1}, but with heavy traffic could be up to 10-100 times greater. The peak measurement in distribution centre had the highest ethylene concentration, as more hydraulic lifts and more trunks and cars parked near the air sampling area.

2.4.2 In-store Ethylene Concentrations

As the result, the chiller rooms in all the locations were set as around 4℃ and around 98% RH (Table 2.10). 0 - 4℃ was recommended as the storage temperature for most fresh (Zhan et al., 2012).

Table 2.10 The average temperature and humidity of the chiller room in different handling area.

Survey site	Temperature (℃)	Humidity (%)
Distribution centre	4.50	97.9
Supermarket A	4	98
Supermarket B	4.16	98.07
Supermarket C	4.23	97.69
Supermarket D	4.11	99.44
Flower distributor	4.21	97.36
Average	4.17	98.05

The 80^{th} percentile ethylene concentration ranged from 116 to 964.5 nL L^{-1} in the distribution centre. For supermarkets, the 80^{th} percentile ethylene concentration in supermarkets as mainly below 50 nL L^{-1}, but the 80^{th} percentile ethylene in supermarket A and supermarket B were 84 nL L^{-1} and 207 nL L^{-1} respectively (Table 2.9). Wills et al. (2000), they previously found ethylene concentrations in supermarkets to be largely between 15 to 100 nL L^{-1}, with only two supermarkets having more than 100 nL L^{-1}.

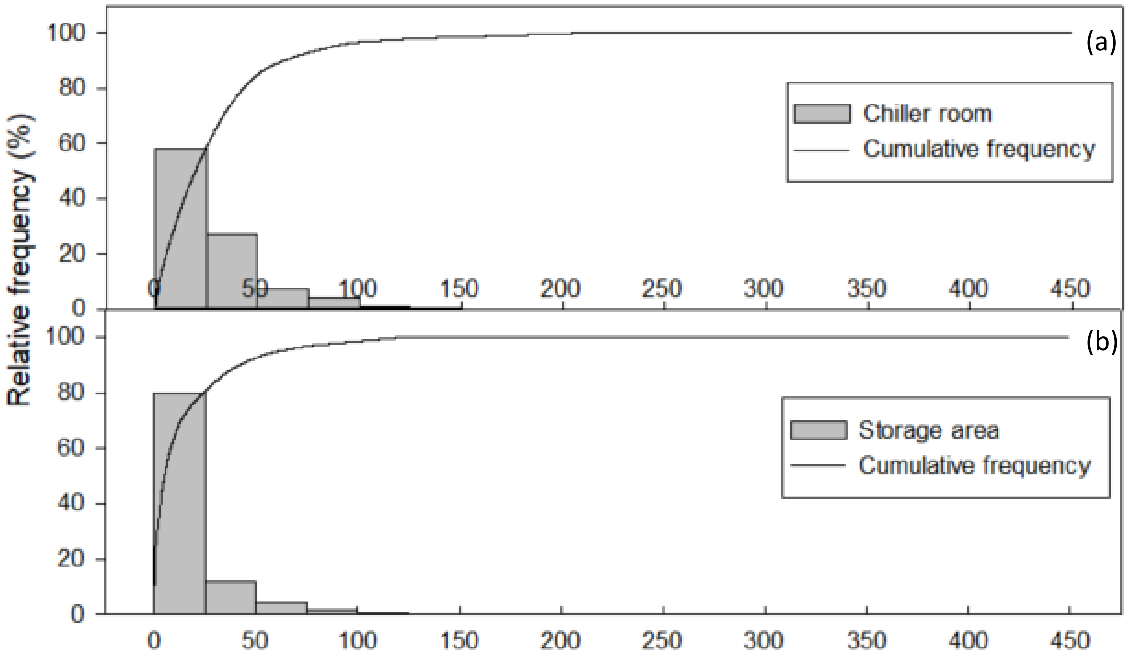


Figure 2.26 Histogram bar and cumulative frequency line of ethylene concentrations measured in different in-store areas at the supermarkets. Locations are (a) chiller room and (b) storage area. Each location contains 3 data points over a 45 min/h period (15min per reading).

According to Figure 2.26, the ethylene concentration of chiller room in supermarkets ranged from 0 to 464.1 nL L^{-1}, while that in storage area ranged from 0 to 361.4 nL L^{-1}. Meanwhile, the 80% ethylene in chiller room and storage area of the supermarkets were 44 and 26 respectively. Rees et al. (2011) found ethylene concentration in supermarkets to range from 0 to 48 nL L^{-1}, was similar to the most supermarkets that were measured. Similarly, Lawton (1991) measured ethylene concentration in supermarket from 17 to 35 nL L^{-1}. Many apples were stored in supermarket A and B was the reason why these two locations had a higher ethylene concentration. One study claimed that the atmosphere with apple can lead to ethylene accumulation (Truter & Combrink, 1994). Furthermore, the peak measured in supermarket C and the chiller room of supermarket A was higher than the peak measured in other supermarkets (more than 300 nL L^{-1}). There were a lot of hydraulic lifts passed in the in-store area was the reason.

According to the results, 80^{th} percentile ethylene concentration of chiller rooms in distribution centre were below 964.5 nL L^{-1}, but the peak measured can be more than 3000 nL L^{-1}. Wills et al. (2000) monitored 35 distribution centres' in-store area and found that 60%

distribution centres had high ethylene concentrations more than 100 nL L^{-1}. Similarly, Rees et al. (2011) estimated that the ethylene concentration in distribution centre without apples ranged from 72 to 548 nL L^{-1}, while ethylene concentrations in distribution centre with apples can reach 3612 nL L^{-1}. The ethylene concentrations in the distribution centre ranged wider than what was observed in the supermarkets. The distribution centre had more petrol exhausted machines than at other places can be a reason. Combustion products can increase ethylene by more than 1000 nL L^{-1} (Keller et al., 2013). Moreover, the products were packaged in the distribution centre could be another possibility, as the fresh inside packages can have ethylene up to 48000 nL L^{-1} (Lawton,1991). There were more products in the distribution centre than supermarkets can be another reason why the ethylene concentration ranged wider in the distribution centre.

Chiller room 5 and 6 in the distribution centre were used to store avocados and apples respectively. The results of this work found the instore rooms with high ethylene production products had higher ethylene concentration than others, as the peak measures in chiller room 5 and 6 were more than 3000 nL L^{-1}. Apple is a high ethylene production product, it can evolve ethylene at a high rate (Wills et al., 2000; Smith et al., 2009). This can account for why the chiller room 6 with apples can have a high ethylene concentration. The main products in chiller room 5 in the distribution centre were citrus. However, citrus can produce very little ethylene (< 0.1 μL kg^{-1} h^{-1}) has been reported by several research studies (Ludford, 2003; Wills et al., 2001; Smith et al., 2009). Avocado and peaches also are high ethylene producing products can account for the high ethylene concentration in chiller room 5 in the distribution centre (Wills et al., 2000; Janssen et al., 2013). Keller et al. (2013) measured the in-store ethylene with apples and found a range from 1.6×10^5 to 5.5×10^5 nL L^{-1}.

Flowers are very sensitive to ethylene in the floral trade. Flowers that respond to low ethylene concentrations are probably those in which ethylene is naturally involved in senescence (Reid & Wu, 1992). As shown in Table 2.9., the 80^{th} percentile in flower distributor ranged 1.3 – 3.8 nL L^{-1} and the peak measured was below 10 nL L^{-1}. The flower distributor can provide a good environment for flowers, considering Reid and Wu found that 10nL L^{-1} ethylene level can cause the reduction in vase life of most cultivars reach around 17%, 38% reduction of *Sandra* and 42% reduction of *Chinera*. Moreover, 100nL L^{-1} can cause around 50% reduction of life of these three flowers, while 1000nL L^{-1} can cause almost 100% reduction in their vase life (Reid & Wu,1992). *Eustoma* flowers did not indicate any morphological changes significantly until the ethylene level reach 1000nL L^{-1} (Ichimura, Shimamura & Hisamatsu, 1998). The flower vase life seems to be not affected by the ethylene concentration in the flower distributor due to our results. The ethylene concentration in flower distributor in our study was extremely low. Ethylene was mainly produced in the pistil of flowers, in particular in the style can be the reason why the ethylene production was similar to ambient (Ichimura, Shimamura & Hisamatsu, 1998). Additionally, there was no air pollution in the flower distributor can be another reason, as the flowers were delivered by labour rather than hydraulic lifts. There are not many studies focus on analysing the ethylene production of flowers. However, it seems that the ethylene production could be involved in several phases of flowers and flowers can be more sensitive during the process of senescence (Ichimura, Shimamura & Hisamatsu, 1998; Reid & Wu, 1992). Serek et al. (2006) claimed that the fourth day of coloration in Orchid *Cymbidium* lips can produce 0nL L^{-1}, while the 16 days can produce 10nL L^{-1} ethylene. Therefore, it is meaningful to gain data from this trial.

From our study, it seems that the most in-store areas provided a good environment to apples, citrus and banana, considering they were not sensitive to the ethylene level below 1000nL L^{-1} (Rees et al., 2011). However, distribution centre, supermarket A and B stored kiwifruit with other ethylene production products can be a risk, as kiwifruit is can be sensitive to ethylene even below 10nL L^{-1} (Jubbar & East, 2016; Rees et al., 2011; Retamales & Campos, 1997). The kiwifruit stored in the chiller room can be ripening very fast, considering its sensitivity (Janssen et al., 2013). It is better to store kiwifruit separately and it was not good to store kiwifruits with the high ethylene production together.

2.5 Conclusion

The data from this part of the experiment indicated that ambient area always had the lowest ethylene level, which was below 100nL L^{-1}. However, the human activities could cause high ethylene levels in the ambient, such as smoking, the gas exhausts from industries and vehicles. The ethylene level of different areas in the distribution, flower distributor and supermarkets were decided by the number of products and the way how did they manage. Basically, the door closed can make the increase of ethylene during the night, as the gas cannot exchange with outside and ethylene can build up. At the same time, the opening door can cause a slight decrease of ethylene. Each supply chain had a different work time cause the difference in the ethylene level.

Overall, the flower distributor did not produce much ethylene, and the level of ethylene was similar to the ambient area. The ethylene level in the flower distributor were below 10nL

L^{-1}, which was extremely low. This is because the flower cannot produce as much ethylene as fruits and vegetables.

Moreover, the ethylene level of chiller rooms in distribution centre ranged widely than other types of supply chain. The reason is that the distribution centre had a huge amount of area and it had more transports to deliver products both inside and outside. chiller room with citrus, kiwifruit, avocado and peaches had the highest ethylene concentration which ranged from 0nL L^{-1} to above 3000nL L^{-1}. Chiller room filled with the apples was the second high, as apple was the high ethylene production fruits. The chiller room with mushroom, root crops and greens similar, which the ethylene level was below 200nL L^{-1} generally.

The ethylene concentration in supermarkets was much lower than the distribution centre is that distribution centres always had more products than the supermarket and the lifting was used more frequency.

Moreover, the way to store products decide the level of ethylene. The ethylene level of chiller room and display area had similar level (below 175nL L^{-1}) of ethylene in Supermarket A, as the products stored in these two locations had a similar amount. Meanwhile, the banana area and shelf area in supermarket B were similar, which were below 175nL L^{-1} as well and they share the same room. The ethylene in chiller room which ranged from 0nL L^{-1} to 400nL L^{-1} in supermarket B. Supermarket C the ethylene level of ambient area, storage area and chiller room were quite similar. This was different from the other two supermarkets. The reason is that the way they stored products was quite different. The products were quite fresh, and they were stored in the chiller room or storage area temporary and then they all be delivered in shopping area. Therefore, the ethylene level in these three areas in supermarket C are similar.

Therefore, it can be concluded from this study that the distribution centre had highest and widest ethylene concentration than other types of supply chain, while the flower distributes had the lowest which was similar with the ambient area. Usually, the ethylene level in supermarkets was below 400nL L^{-1}, and the ethylene level depend on the way they manage the products. Nonventilated environment can cause the build-up of ethylene. Besides, the temperature and humidity of most supply chain was set as around 4℃.

Moreover, so many places, the keeper prefer put kiwifruit with other high ethylene production products, this can be a risk. As kiwifruit is a high ethylene sensitive product. It is better to store kiwifruit with low ethylene production products or individually.

Chapter 3 Broccoli Responses to Exogenous Ethylene Environments

3.1 Introduction

Previous chapter gives us values of concentrations and the temporal nature of these concentrations. However, these are somewhat meaningless without knowledge of how the product responds to these concentrations. Each product responds differently, and hence quantitative documentation of ethylene dose impact a fairly monumental task. Websites such as UCDAvis produce facts provide excellent guidelines. This work chooses broccoli as an example case study how to quantify potential quality effects of environmental ethylene.

Broccoli (*Brassica oleracea* var. *italica*) is a common and popular vegetable in the market all around the world. Broccoli contains a wide range of phytochemicals (e.g. gluosinolates, flavonols, and carotenoids), vitamins and sugars (Jones et al., 2006). However, the shelf life of broccoli is short, as it is a rapidly developing floral vegetable (Forney, 1995; Tian et al., 1995). Esturk et al. (2014) claimed that broccoli can store around 20 days at 4 ℃.

Colour changes and senescence are the main causes of losses during the postharvest period (Forney, 1995; Jones et al., 2006; Tian et al., 1995; Tian et al., 1996). The green colour is a significant commercial quality index of broccoli (Gong & Mattheis, 2003), with degreening limiting market acceptance (Tian et al., 1996). Degradation of chlorophyll is the main cause of the yellowing of broccoli (Cai et al., 2019). The degradation of chlorophyll can cause colour changes, such as yellowing in the green plants (Able et al., 2003; Lu, 2007; Sledz et al., 2016) and can be used as an index to evaluate the senescence (Wu et al., 2019). Colour also can be a factor that impacts on the choice of customer (Sledz et al., 2016) with the yellowing of greens leading to consumer rejection (Kaiser et al., 2012). Moreover, colour can pose a primary role in the taste of sweetness and pleasantness (Wu et al., 2019). Clydescale (1993) found that the green-coloured products were truly sweeter than colourless products.

Able et al. (2005) noted that ethylene has a significant role in chlorophyll degradation. Tian et al. (1995) provided evidence that ethylene plays a significant role in regulating chlorophyll loss in broccoli and hence considered as a highly ethylene sensitive product. Gong and Mattheis (2003) treated broccoli with 10^6 nL L-

^{1} for 5 hours at 20℃ and they found the colour changes (presented as hue angle) of broccoli with ethylene treatment was the yellowest (h= around 92), while the broccoli with 1000 nL L^{-1} 1-MCP treatment (h = around 117) inhibited the yellowing process. Similarly, broccoli with 1000 nL L^{-1} ethylene treatment at 10℃ (h = around 90) yellower than other treatment, while the broccoli treated with 1000 nL L^{-1} 1-MCP (h = around 123) inhibited the yellowing process (Fan & Mattheis, 2000).

Colour change and weight loss are two quality attributes to assess the quality changes of broccoli during shelf life. Water loss can cause decay, shrivel development and weight loss during the storage of green products (Soysal, 2004; Lu, 2007). Tian et al. (1995) found that the large florets of broccoli lost water at a faster rate than smaller or medium sized florets. Perini et al. (2017) demonstrated that the weight loss of broccoli at 20℃ for three days were between 3-4%. Additionally, 2.17% (light conditions) and 1.95% (dark conditions) weight loss were found separately after 10 days with 7℃ (Zhan et al., 2012). Raffaella et al. (2010) treated broccoli with different temperature for 17 days, and they found that broccoli with 5℃ had 6.8% weight loss and broccoli with 10℃ can be more than 14%. The broccoli with 4 ℃ for 6 days had 5.51% weight loss had been found by Nath et al. (2011)'s study. Similarly, Gomes et al. (2008) demonstrated that the weight loss of broccoli with 4℃ increased during the storage and the weight loss after 14 days was 21.75%.

It is shown that broccoli can be sensitive to the critical ethylene concentration which is above < 100 nL L^{-1} (Rees et al.2000). Similarly, Ku and Wills (1999) provided evidence that reduction ethylene concentration (from 100 nL L^{-1} to 5 nL L^{-1}) can double the shelf life of broccoli. According to the results from chapter 2, 80th percentile of ethylene concentration of chiller rooms in distribution centre were more than 100 nL L^{-1}. For supermarkets, although the 80th percentiles of most areas were below 100 nL L^{-1}, the peaked measured can be more than 100 nL $L^{-1.}$ Hence, the measured concentrations are likely to have a detrimental effect on broccoli in the supply chain. The objectives of this work were to quantify the detrimental effects (if any) of the ethylene concentrations measured in real storage conditions on broccoli. With this information it can be determined whether the concentrations measured in the supply chain potentially contribute to reduced shelf life of broccoli, and hence whether remedial action may be worthwhile.

3.2 Methodology and materials

3.2.1 Plant material

A total of 300 broccoli (*Brassica olerace* var. *italica*) were sourced from Woodhaven Gardens, Levin and were sent to the Postharvest Laboratory, Massey University Palmerston North. Every 25 broccoli were packaged to one box, and the boxes were held by plastic bags. The temperature of transportation conditions between harvest and delivery was 0℃, and the time between harvest and delivery was around 1 hour. A total of 150 broccoli were used for a two-week trial (Figure 3.2), with the trial being repeated. After the arriving of broccoli, randomization was conducted, and then groups of 5 broccoli was put into a mesh bag. A total 30 bags for one trial were required.

3.2.2 Ethylene continuous flow system

An ethylene flow-through system was established in a temperature-controlled room (TCR). Referring to the temperature and humidity information measured from most supply chain areas surveyed (refer to Table 2.9) the room was set at 4 ℃ and 98% RH.

With reference to the survey data (Table 2.10) collected in the previous chapter, 5 ethylene treatments of 0, 50, 100, 500 and 1000 nL L^{-1} were chosen. Generally, 0 nL L^{-1} was chosen to imitate the ambient environment. 50 nL L^{-1} was chosen, as the 80th percentile ethylene of storage area in supermarkets was around this level. 80th percentile ethylene concentration of most chiller rooms in distribution centre and peaked measured in supermarkets were around 100 and 500 nL L^{-1}. On occasions, the ethylene concentration was found to be more than 1000 nL L^{-1} in the distribution centre due to the peaked measured.

A continuous flow system was established to deliver the desired ethylene gas concentrations to the broccoli (Figure 3.1). Dry air, 10^5 nL L^{-1} and 4500 nL L^{-1} ethylene standards were used to achieve the desired ethylene concentrations. The 0 nL L^{-1} treatment was only connected with dry air. The 50 nL L^{-1} and 100 nL L^{-1} ethylene treatment was created through dilution of the 4500 nL L^{-1} ethylene standard. Similarly, 500 nL L^{-1} and 1000 nL L^{-1}, the 10^5 nL L^{-1} ethylene standard was diluted with dry air. Mass flow controllers (GSC-B9TABB23/21, Vögtlin Instruments GmbH, Aesch, Switzerland) were utilized to adjust the flows of the air and ethylene standard in order to obtain the desired concentrations of ethylene in the resulting mix.

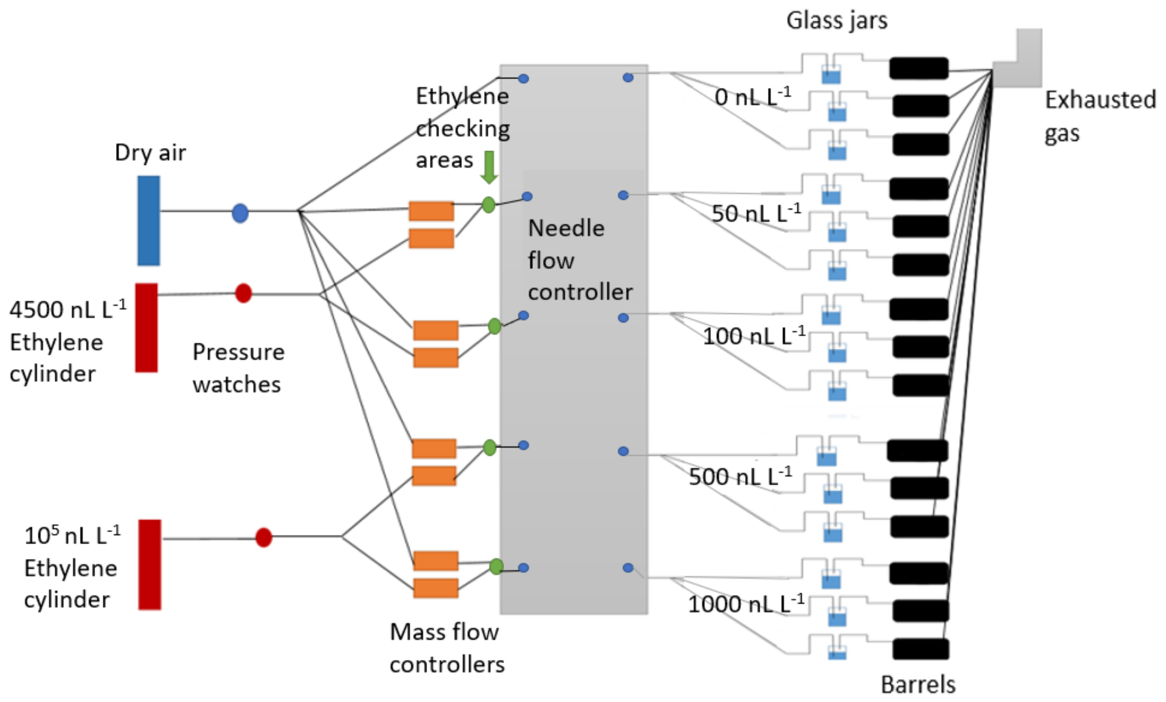


Figure 3.1 The layout of the ethylene continuous flow system.

After mixing at 20 °C, tubes of each gas mix were connected into a needle valve flow controller in the temperature-controlled room, where each concentration was divided equally into 3 branches. The needle valve flow controller was used to ensure an equal flow rate of 230 $mL.min^{-1}$ through each PVC container. Flow rates were checked with MACView twice a week. The resulting 15 tubes, (3 of each concentration) connected each to one of 15 PVC containers (in 6 litres), each containing 2 bags of broccoli. Prior to entering the PVC containers each gas flow was bubbled into a 10% glycerol in water mixture (2000 mL) to check the leakage. Outflows from each container were connected to a room venting systems to minimise any cross contamination of treatments.

3.2.3 Ethylene concentration checking

In this section, the ethylene level will be checked twice a week to ensure the ethylene treatment is stable and correct. All the data and the information has been showed in the table below. At the beginning, the MACView connect with 970ppb ethylene standard and 92ppb standard to conduct the calibration. The MACView got two readings, which were 990ppb and 90ppb. Then set reading from the MACView as x, the standard as y, put them into the formulation y=kx+b and get the calculation. This process has been showed below.

$$\begin{cases} 970=990k+b \\ 92=90k+b \end{cases}$$

After the calculation, the value of k and b has been gained, which are 0.9756 and 4.2585 respectively:

$$\begin{cases} k=0.9756 \\ b=4.2585 \end{cases}$$

Therefore, the actual ethylene level formulation has been gained which is y=0.9756x+4.2585. After getting readings from MACView, the readings should be put into this formulation to gain the actual ethylene level. In this way, whether the ethylene level is correct can be evaluated. Usually, MACView can cause 10% errors. Making sure the actual level is around 10% is significant.

As shown in the table, the 500ppb ethylene treatment on 4th December is quite high, so the flow rate of air is increased by 1.1. Generally, the ethylene level is quite stable, and all the errors are within 10%.

Ethylene concentrations within the experiment were checked twice a week. The needle was injected through the tubes, which was located behind the mix, it connected with MACView (Figure 3.1). After getting three readings from one treatment, the needle injected to another until the four ethylene treatments all checked (Table 3.1). 0 nL L^{-1} was not be checked considering it was connected with dry air directly. The flow rates were adjusted until the required ethylene concentration was gained. Ethylene concentrations achieved are showed in Table 3.4. Generally, the ethylene concentration was quite stable, and within 10% of the experimental design concentration.

Table 3.1 The ethylene readings from the regular check and the adjustment of the value of mess flow meter.

Date	Treatment (nL L^{-1})	MACView (nL L^{-1})	Actual ethylene level (nL L^{-1})	Error (%)	Flow rate of air (ml/min)	Flow rate of ethylene(ml/min)
19/11	50	52	55	4	533	8.1
	100	98	100	-2	749	16.5
	500	611	600	22	767	4.1
	1000	968	949	-3	755	7.5
23/11	50	49	52	-2	533	8.1
	100	95	97	-2	749	16.5
	500	525	516	5	767	4.1
	1000	992	972	-1	755	7.5
25/11	50	52	55	4	533	8.1
	100	96	98	-2	749	16.5
	500	461	454	-8	767	4.1
	1000	963	944	-4	755	7.5
26/11	50	46	49	-8	533	8.1
	100	95	97	-2	749	16.5
	500	547	538	9	767	4.1
	1000	996	976	0	755	7.5
04/12	50	52	55	4	533	8.1
	100	101	103	-2	749	16.5
	500	568	558	14	767	4.1
	1000	1053	1032	5	755	7.5
6/12	50	51	54	2	533	8.1
	100	99	101	-2	749	16.5
	500	520	512	4	843	4.1
	1000	955	936	-5	755	7.5
18/12	50	52	5	4	533	8.1
	100	98	100	-2	749	16.5
	500	520	512	4	843	4.1
	1000	1010	990	1	755	7.5
19/12	50	51	54	2	533	8.1
	100	98	100	-2	749	16.5
	500	518	510	4	843	4.1
	1000	1005	985	1	755	7.5

3.2.4 Broccoli quality assessment

Each barrel contained 2 bags of 5 broccoli each. One of these bags was removed out and measured (non-destructively) at on day 0, 4, 8, 11 and 14 of the experiment. Meanwhile, the other bag from each barrel was maintained in the barrels, only being measured at the beginning and the end of storage. In conducting the experiment this way, a check of the effect of removing from the ethylene and temperature control (in order to measure) could be conducted.

Weights of broccoli were measured as singles via electronic balance (3000D SCS, Precisa, Switzerland) on day 0, 4, 8, 11 and 14 at 4℃. The equipment was fully covered by the plastic bag to handle condensation before moving to the chiller room. The percentage of CO_2 and O_2 of each barrel was measured by gas analyser (Analytical Development Company, Hoddeston, UK) on day 2, 7, 11, 14 to gain the respiration rate.

Berset & Caniaux (2019) illustrated that the colour is nonuniformly distributed over the surface of the food. Colour changes of broccoli were measured using a spectrophotometer (CM-2600d, Konica Minolta, Japan) at 4℃. Results were reported in the CIELab colour space descriptors of C (chroma), h (hue angle), L (lightness from black to white), a (redness/greenness), b (yellowness/blueness). Three (3) locations of the head area of broccoli were measured (Figure 3.2) and averaged.

Additionally, visual colour changes were also recorded by photography (both side and head area of broccolis). All pictures were taken with a Canon EOS 600D, with the exposure time set to 1/160 sec and ISO speed of 400. Pictures were captured within a lightbox fitted with D65 lighting that was turned on at least 10 mins earlier to ensure stable pictures capture.

Figure 3.2 The layout of the measurement locations (the head of broccoli).

3.2.4 Data Analysis

Data was collated in Excel (64-bit Version, Microsoft, USA) and significant differences in means determined by conducting an ANOVA analysis by Minitab (Version 19.0, Statistical software, USA). Data is presented by plotting with Sigmaplot (Version 14.0, Graph software, USA).

3.3 Results and Discussion

3.3.1 Weight loss

The weight loss of broccoli is not due to solely water loss, but also due to the fluorescence abscission. As the florets of broccoli can drop down due to the senescence and this phenomenon can affect the weight loss. Weight loss was influence by ethylene through induction of fluorescence abscission, in addition to weight loss by water loss. As a result, weight loss was higher in the higher ethylene treatments and there was a significant difference between the weight loss with high ethylene concentration (500 and 1000 nL L^{-1}) and low ethylene concentration (0 and 50 nL L^{-1}). Moreover, the weight loss of repeat measures groups was higher than the weight loss of single measure group. Handling the broccoli in the experiment created further weight loss as handling caused additional damage to the florets, causing more floret drop.

Overall, the weight loss increased during the storage time with the largest weight loss (1000 nL L^{-1}) of repeat measures groups being approximately 5% over 14 days, a rate of 0.36 $\%.day^{-1}$ (Figure 3.3a-b). The weight loss of 500 nL L^{-1} was the next highest in repeat measures groups, which was around 4%. In addition, the weight loss of 0 nL L^{-1} and 50 nL L^{-1} had a similar trend of ethylene breaking down treatment in trial B, and they reached around 2% on the last day. It was around 1% lower than 100 nL L^{-1}. However, the weight loss of 0 nL L^{-1} of repeat measure group had a similar trend as 100 nL L^{-1} in trial A, and it was 3.4% at the last day. The weight loss with 50 nL L^{-1} was the lowest in trial A.

For the single measure groups (Figure 3.3c-d), the weight loss increased during the storage days as well. Higher ethylene treatment had higher weight loss, due to the fluorescence abscission. The weight loss of 1000 nL L^{-1} of control groups in trial A and trial B were 2.4% and 4% separately. 50 nL L^{-1} had the lowest weight loss (being around 1%)

at the last day, while the weight loss of 0 nL L^{-1} was around 0.5% higher than the weight loss of 50 nL L^{-1}. However, there was no significant difference between them. The weight loss of 100 nL L^{-1} was the 3rd high was between high ethylene concentration (500 nL L^{-1} and 1000 nL L^{-1}).

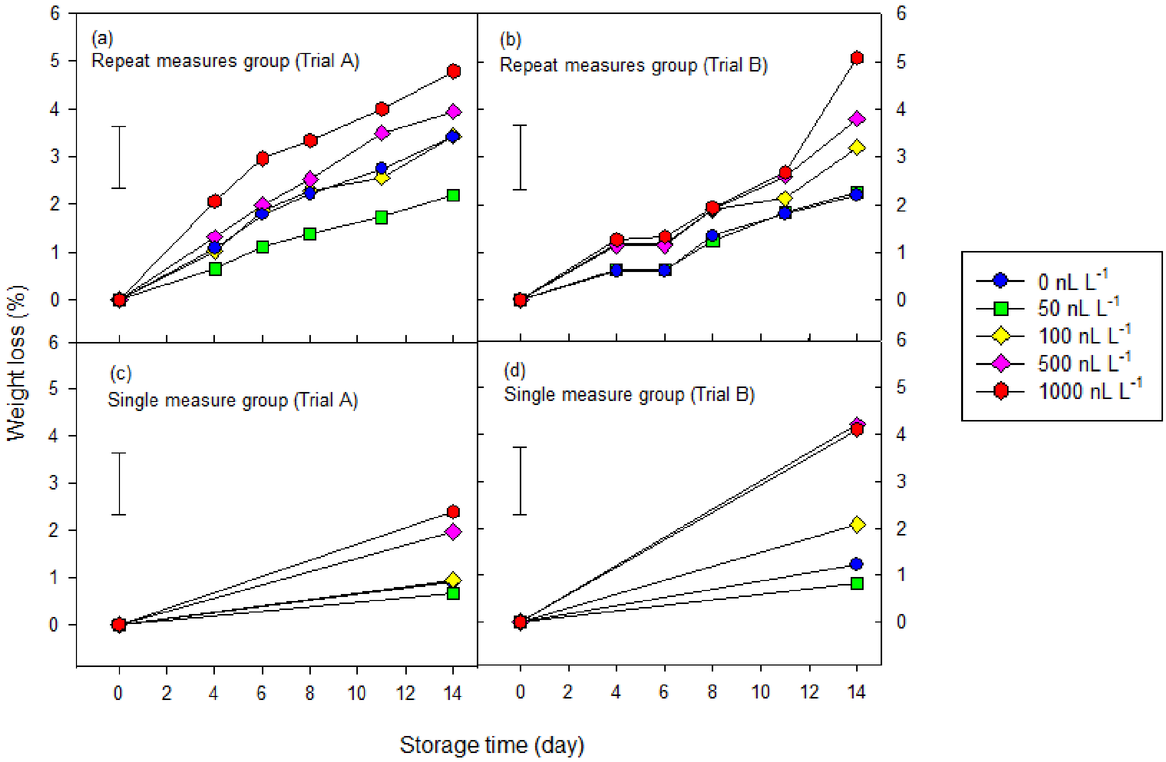


Figure 3.3 Weight loss during storage for broccoli exposed to exogenous ethylene, while at 4℃ and 98% RH. (a-b) repeated measures groups. (c-d) single measures group. Data points are averages of 30 broccoli. LSD0.05 has been shown as error bars.

3.3.2 Respiration rate

For the respiration rate, there was not an obvious increase trend during the storage (Figure 3.4a - b). Although the higher ethylene concentration had a higher respiration rate but there was no significant difference between each treatment. The barrels opened to pull out the broccoli during the measurements cause the unstable O_2 and CO_2 rate. Therefore, the result of respiration rate was not regular. For trial A (Figure 3.4a), 1000 nL L^{-1} had the highest respiration rate which was around 44 mg(μl)/(h·g). The highest respiration rate of trial B (Figure 3.4b) was 500 nL L^{-1} at the end (about 48mg(μl)/(h·g)), which was similar to the respiration rate of 1000 nL L^{-1} (About 44 mg(μl)/(h·g)). 0 and 50 nL L^{-1} had the lowest respiration rate in both trials, which was around 23 mg(μl)/(h·g).

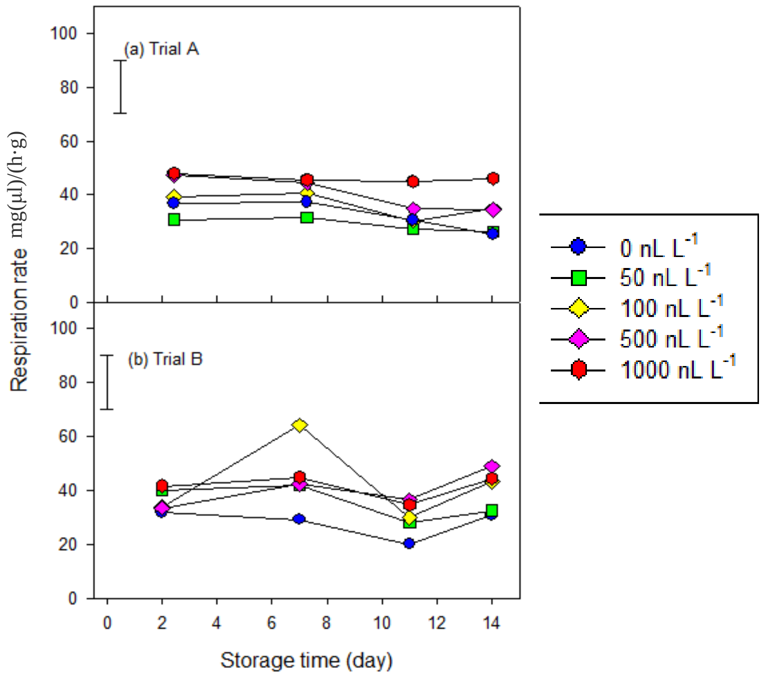


Figure 3.4 Weight loss during storage for broccoli exposed to exogenous ethylene, while at 4℃ and 98% RH. (a) Trial A. (B) Trial B. Data points are averages of 30 broccoli. LSD0.05 has been shown as error bars.

3.3.3 Colour changes

3.3.3.1 Chroma (C value)

The Chroma value tended to increase during the storage. Moreover, the broccolis with 1000 nL L^{-1} ethylene had the highest C value at the end, while 50 nL L^{-1} treatment had the lowest C value.

For both repeat measures groups, 1000 and 500 nL L^{-1} had a similar level of C value at the end which was around 24 (Figure 3.5a - b). At the end, the broccoli with 100 nL L^{-1} treatment had similar C value (at 23) as 0 nL L^{-1}. The broccoli from repeat measures group of trial A treated with 50 nL L^{-1} had the lowest C value, which was around 18 nL L^{-1} (Figure 3.5a). For ethylene repeat measures group of trial B (Figure 3.5b), the 1000 nL L^{-1} ethylene treatment increased from 13 to 23, which had the highest C value at the end. It was followed by 500 nL L^{-1}, which was around 1 unite lower than 1000 nL L^{-1}. The C value of 100 and 0 nL L^{-1} had A similar C value, but 0 nL L^{-1} had a higher C value (18) at the end. The 50 nL L^{-1} treatment had the lowest C value at the end which was at 17.

For single measure group of trial A (Figure 3.5c), the C value of broccoli with 1000 nL L^{-1} treatment reached around 28 at the end, which was as around twice as the C value level of 0 and 50 nL L^{-1} treatment. The C value of 1000 nL L^{-1} treatment was 10 units lower than that of 500 nL L^{-1} treatment. For the single measure group of Trial B, the C value increased slightly during the increase of storage time as well (Figure 3.5d). The 1000 nL L^{-1} had the highest C value at the end (around 28), while 0 nL L^{-1} had the lowest C value (around 18). It seems that the higher ethylene treatment had the higher C value.

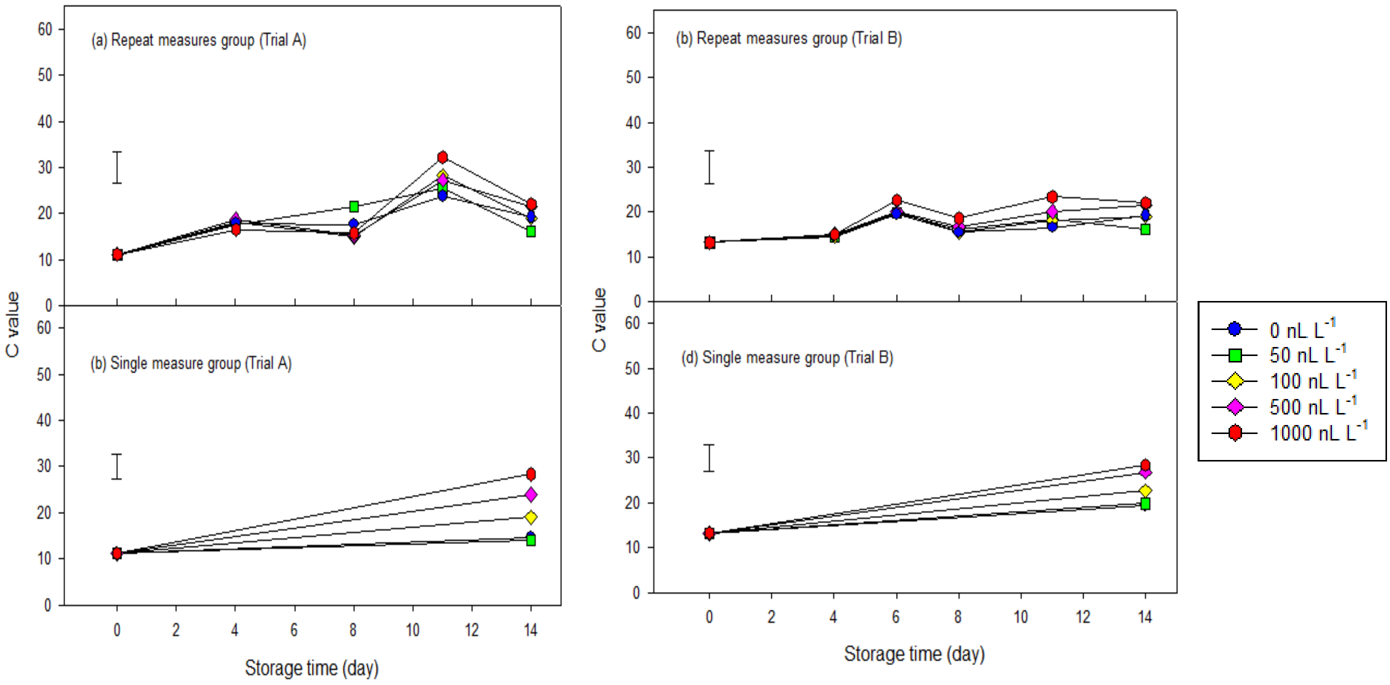


Figure 3.5 C value during storage days of broccolis ethylene breaking down treatment and control group in trial 1 and Trial B at 4℃ and 98% RH. (a-b) repeated measures groups (As there was a calibration error of the colour measurements on day 6 of repeat measures group in trial A, the data was deleted from the graphs.).(c-d) single measures group. Data points are averages of 30 broccoli. LSD0.05 has been shown as error bars.

3.3.3.2 Hue angle (h value)

Overall, the hue angle value had a decrease trend from all the results. There was a negative correlation between h value and storage time of both repeat measures groups and single measure groups as well. The significant difference was shown around day

12. From Figure 3.6, all the groups with 1000 nL L^{-1} treatment had the lowest hue angle at the end, which were around 95° in repeat measures groups (Figure 3.6a - b) and 91° in single measure groups (Figure 3.6c - d) respectively. Meanwhile, the control and 50 nL L^{-1} from the experiment had the similar highest H value (around 115). The h value with 100 nL L^{-1} was between the h value of 500 and 50 nL L^{-1}. It seems that the higher ethylene treatment had the lower h value due to the information of the graph, and there was not a significant difference between control and 50 nL L^{-1}.

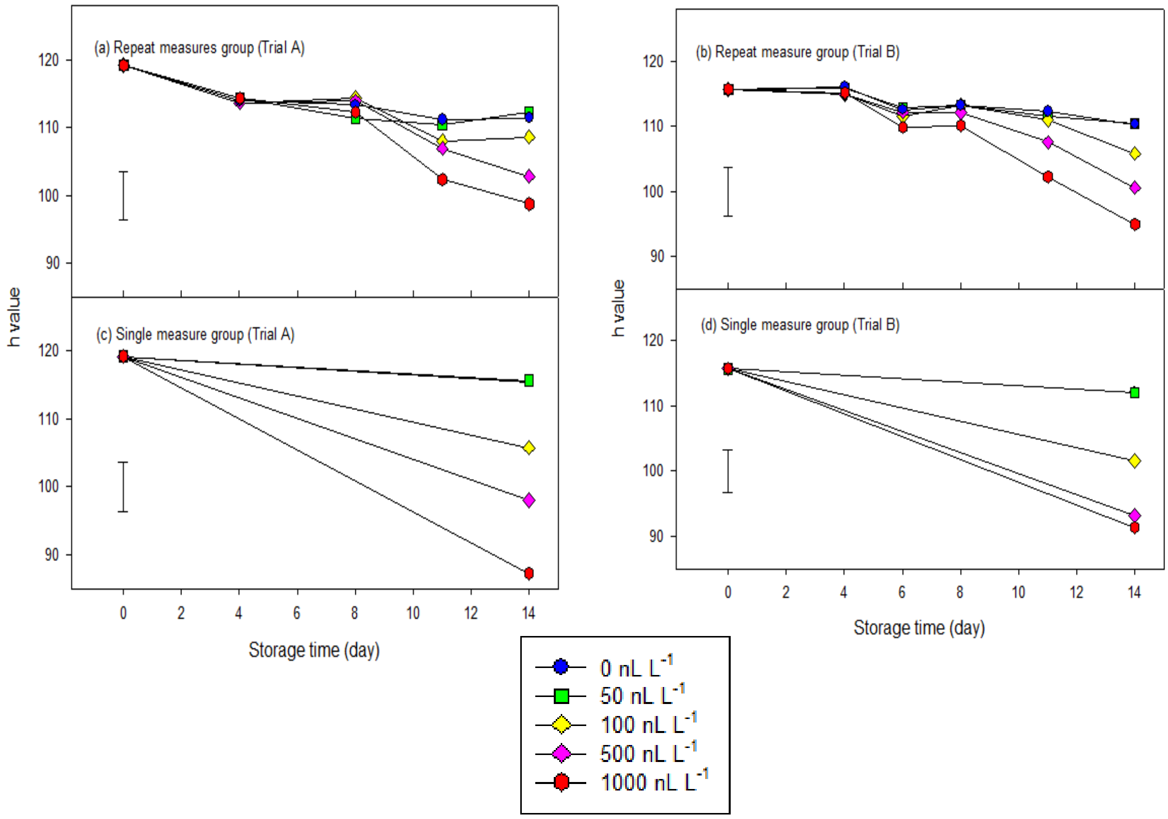


Figure 3.6 H value during storage days of broccolis ethylene breaking down treatment and control group in trial 1 and Trial B at 4℃ and 98% RH. (a-b) repeated measures groups (As there was a calibration error of the colour measurements on day 6 of repeat measures group in trial A, the data was deleted from the graphs.). (c-d) single measures group. Data points are averages of 30 broccoli. LSD0.05 has been shown as error bars.

3.3.3.3 Lightness (L value)

Generally, if there was a change in lightness, there was an increase in lightness value during the experiment (Figure 3.7). Lightness started at 39.8. Differences between ethylene concentrations tended to develop after only 4 days storage (the first point of the measure was 39.8 in the experiment).

For repeat measure groups, at this time the 1000 and 500 nL L^{-1} treatments had differentiated from the others, suggesting that even small time periods to high ethylene exposure can at least quantifiable reduce broccoli storage life (Figure 3.7a-b). Moreover, the L value of 500 and 1000 nL L^{-1} was significantly different from 0 and 50 nL L^{-1} ethylene treatment. By the completion of the trial, the 1000 nL L^{-1} treatment had the highest lightness value (approx. 45). Meanwhile, the control and 50 nL L^{-1} treatments remained at similar values as at the start of the experiment (e.g. approximately 40) and there was not a significant difference between these two treatments. Moreover, the L value of 100 nL L^{-1} (around 40) was between it the L value of 100 and 500 nL L^{-1} ethylene treatment. The repeated measures group tended to have slightly lower responses to the ethylene treatments than the single measures group, when comparing the measured values at 14 days. For the single measure group, ethylene concentration can be more than 50 in high ethylene treatment (500 and 1000 nL L^{-1}) (Figure 3.7c-d). This is due to the single measure groups was exposure to ethylene a longer time than repeat measures groups.

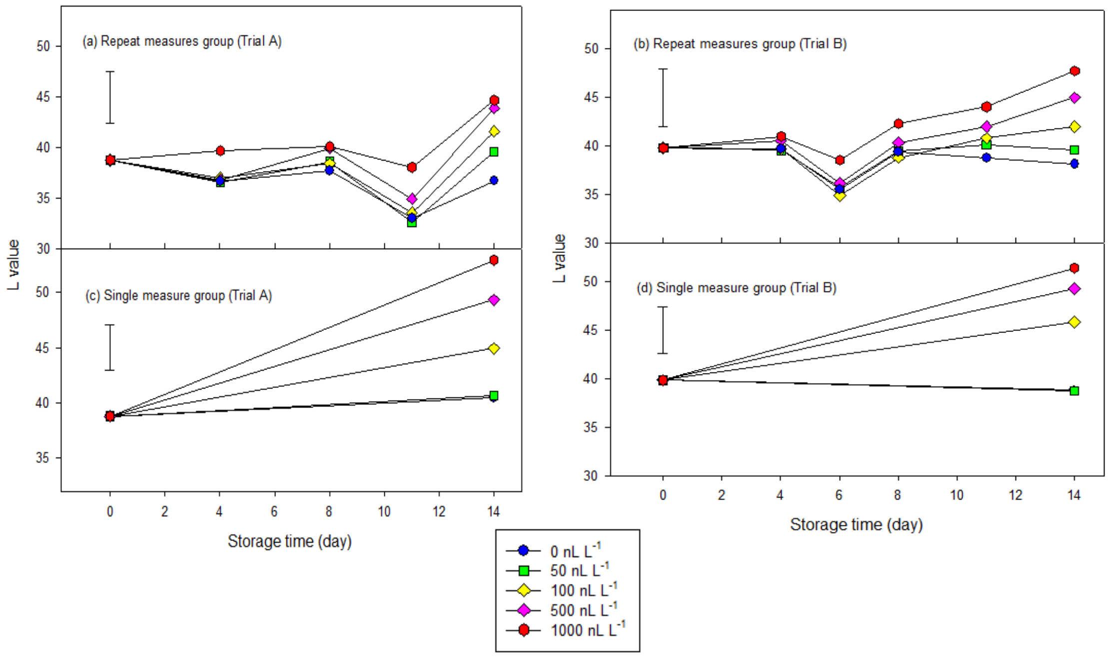
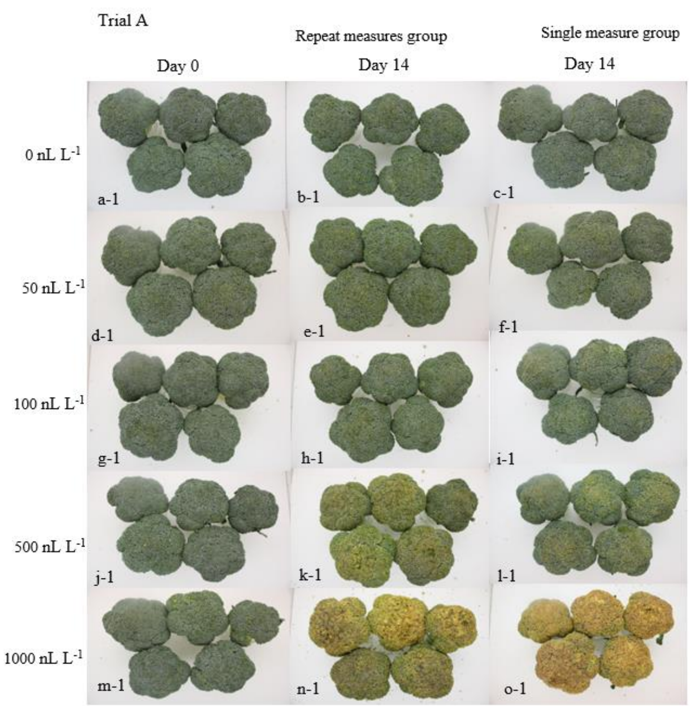


Figure 3.7 Lightness value during storage of broccoli exposed to exogenous ethylene at 4℃ and 98% RH. breaking down treatment and control group in trial 1 and Trial B. (a-b) repeated measures groups(As there was a calibration error of the colour measurements on day 6 of repeat measures group in trial A, the data was deleted from the graphs.). (c-d) single measures group. Data points are averages of 30 broccoli. LSD0.05 has been shown as error bars.

3.3.4 Visual colour changes

The broccoli reduced in size and became yellower during the storage time (Figure 3.8). Under continuous ethylene treatment, the visual colour changes were not obvious immediately obvious to the eye in low ethylene concentration (< 100 nL L^{-1}) treatments. With > 500 nL L^{-1} for 14 days ethylene yellowing, and fluorescence abscission was obvious. For the colour measurement, the C, h, L value dropped dramatically can account for the obvious visual changes. Furthermore, there was no obvious colour changes of 0, 50 and 100 nL L^{-1}, as the C, h, L value was not a significant difference. There were also some black areas of the broccoli heads, especially in the repeated measures group, suggesting that this type of injury may be a result of physical injury caused by the multiple handling occasions. Yellowing of the single measure group is observed to be potentially more severe than for the repeated measures group, potentially as a result of not be removed from ethylene for measurement.

The florets of broccoli from high ethylene treatment (> 500 nL L^{-1}) dropped down seriously when they were taken us for the photography. This associated with the high weight loss in high ethylene concentration than low ethylene concentration.



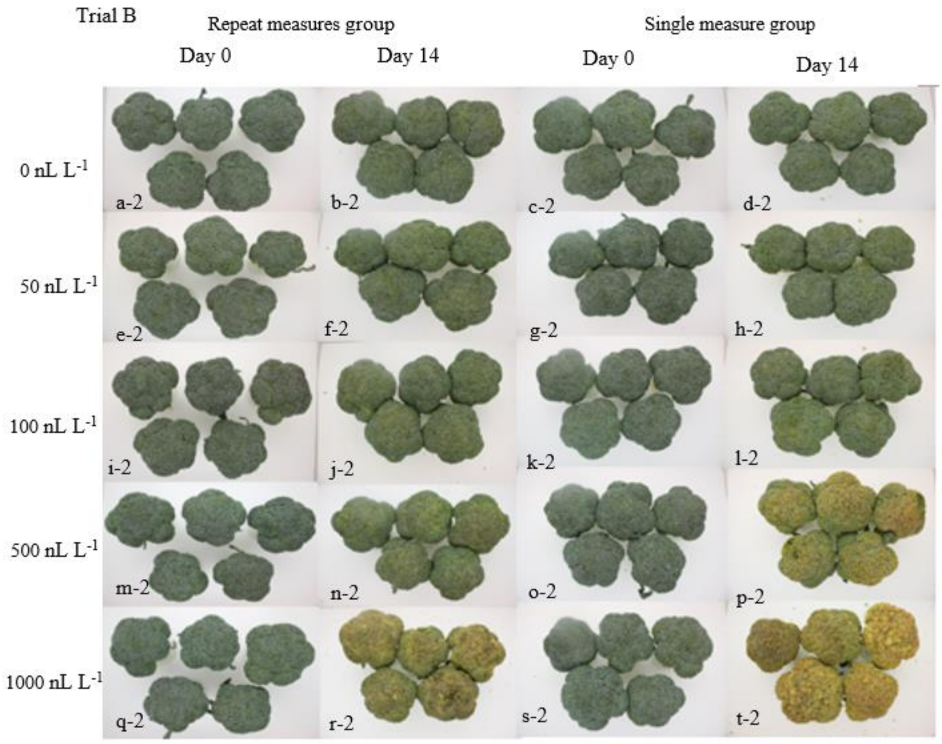


Figure 3.8 Visual observation of broccoli from head area of ethylene breaking down treatment and control group between the first day and last day of the first trial in trial A and trial B

3.4 Over Discussion

3.4.1 Weight loss

From our result, the weight loss of broccoli with 4℃ and 98% RH for 14 days was all below 10 %, and the broccoli with high ethylene concentration had around 5 % weight loss. This is similar to Raffaella et al. (2011)'s study, as they found the broccoli with 5℃ for 17 days had 6.8% weight loss. Besides, the weight loss of broccoli for 6 days with 4℃ was 5.51% due to the floret's deterioration (Nath et al., 2011). Similarly, Gutierrez-Martinez (2010) treated banana fruits with 0, 10^4 10^5 and 10^6 nL L^{-1} ethylene, and they found the weight loss increased for 10 days period and the weight loss rate was below 10% due to the water loss.

3.4.2 Respiration rate

There is a positive relationship between senescence and respiration (Li et al., 2016; Jiang et al., 2017). In this study, the senescence of broccoli with high ethylene concentration was faster than that with low ethylene concentration. It seems that the higher ethylene concentration had higher respiration rate, but there are not significant difference between each treatment. The barrels opened to pull out the broccoli during the measurements can be the reason why there was no significant difference.

3.4.3 Colour parameters

Due to the result, the chroma (C value) increased during the ripening. 1000 nL L^{-1} ethylene treatment had the most significant effects on C value of broccoli, which the C value dropped fastest. Also, the high ethylene concentration (500 and 1000 nL L^{-1}) had significantly difference from the control group. Additionally, the C value of the control group had a lower C value than ethylene breaking down treatment. Therefore, the continuous ethylene treatment can have the worst effects on broccolis. Similarly, Gutierrez-Martinez (2010) treated banana fruits with 0, 10^4 10^5 and 10^6 nL L^{-1} ethylene, and they found higher ethylene treatment had high C value, and the over trend of C value was increased. In addition, one study estimated that C value was calculated via formulation $C = (a^2 + b^2)^{1/2}$ (Soysal, 2004). a value (Figure A.6a) and b (Figure A.6b) value increased in our results, so C value also had the increase trend. Wu et al. (2019) demonstrates that the increase of b value can correspond to the degradation of chlorophyll, which can cause the yellowing of vegetables. Similarly, Fang et al. (2016) also estimated that the higher b value had a more obvious yellowing phenomenon. Furthermore, another study found the broccolis treated with 1000 nL L^{-1} had a higher value than the control group (McGuire, 1992; Gong & Mattheis, 2003).

Overall, the hue angle (h value) had a declining trend during the storage time. the effects of high ethylene concentration (500 and 1000 nL L^{-1}) on h value was significantly different from 0 and 50 nL L^{-1}. Tian et al. (1994) estimated that the broccoli treated with 50mL 80% ethylene had a lower hue angle than the control group. However, they found there was no significant difference in the effects of h value between control group and ethylene treatment. Their trial was conducted only three days, it may hard to figure out the difference between the control group and ethylene treatment in such a short period of time. Similarly, ethylene with 100 nL L^{-1} had the lowest hue angle with the control group, and the h value of them was significantly different (Cai et al., 2019). Another article also proved that the higher ethylene had a lower h value. Tian et al. (1994) used cytokinin to reduce the effects of ethylene on broccoli. They found higher cytokines cause the lower ethylene production of broccoli, and the h value was higher with lower ethylene production.

Also, the H value of broccoli treated with 10^6 nL L^{-1} ethylene was lower than the control group were reported via Gong and Mattheis (2003).

According to the result of lightness (L value), it seems that the higher ethylene treatment had a higher L value, and L value climbed during the storage. The broccolis florals were dropped down due to the ripening and senescence during the storage, and this phenomenon can cause the increase the lightness. Moreover, 1000 nL L^{-1}had the highest L value at the end. However, 0 nL L^{-1} was not significantly different from 50 nL L^{-1} treatment. Gong and Mattheis (2003) also compared L value between air and 1000 nL L^{-1} ethylene, and they found the ethylene treatment had higher lightness than the control group, and the lightness in the air was significantly different from the lightness with ethylene. This result was similar to our result. However, it seems that there was a decreased of the lightness from first day to last day (Gong & Mattheis, 2003). This is different from our result. However, this can be account for that they only measured L value of broccoli for 3 days. From our result, the lightness from day 0 to day 6 also had a decreased trend.

3.5 Conclusion

From the results, 1000 nL L^{-1} ethylene treatment had the worst effects on broccolis shelf life. At the end of the trial, the yellowing of 1000 nL L^{-1}treatment was more obvious and visually. By comparison, it is hard to see any colour changes of other ethylene treatments through the picture. Moreover, it seems that 0 nL $L^{-1}$and 50 nL L^{-1}had similar effects on the quality of broccolis, considering there did have a significant difference in some indexes. All the results follow a similar role-higher ethylene had worse effects than lower ethylene treatment. During the storage, the weight of broccoli decreased due to the shriving of broccolis. The 1000 nL L^{-1} had the highest weight loss, as the high ethylene concentration speeded up the process of ripening and the floral of broccolis dropped down seriously on the last day of the trial. Therefore, it seems that stored broccolis in the chiller room with higher than 1000 nL L^{-1}ethylene can cause a serious impact on vegetables.

Chapter 4 Discussion and Recommendation

4.1 Industry survey

The ethylene concentration in the supply chain distributor and retail was studied. In the ambient environment, 80% of the measured value was below 10 nL L^{-1}, although some peak values measured of the ambient area were higher than 50 nL L^{-1}, with the ambient area in the distribution centre peaking at 136.2 nL L^{-1}. These peak measures may have been influenced by the local release of anthropogenic sources, such as hydraulic lifts and trucks in the loading area near to the sampling location (Keller et al., 2013).

The ethylene concentration in chiller room of supermarkets ranged from 0 to 464.1 nL L^{-1}, with more than 80% of the measured values below 100 nL L^{-1}. An exception to the measurements was the ethylene concentration of the chiller room in supermarket B, where the 80^{th} percentile value was 207 nL L^{-1}. Supermarket B stored more high ethylene producing products (950kg apples) in the chiller room possibly contributing to the high ethylene concentration observed. The storage area in supermarkets ranged from 0 to 361.4 nL L^{-1}. This is much wider than the findings of Rees et al. (2011) who measured ethylene concentration of storage areas in different supermarkets and reported a range of 0 to 91 nL L^{-1}. Similarly, Wills et al. (2000) reported that only 2% of the storage area in supermarkets had more than 100 nL L^{-1} ethylene concentration.

The ethylene concentration of the rooms in the distribution centre with high ethylene producing products (e.g. apples and avocados) (between 752 and 964.5 nL L^{-1} at 80^{th} percentile) was much higher than the chiller rooms with few ethylene producing products (between 110 and 150 nL L^{-1}).

Rees et al. (2011) reported that below 100 nL L^{-1} ethylene concentration already can affect the postharvest life of horticultural products. Therefore, there is a risk for both supermarkets and distribution centre in the current supply chain, considering the average ethylene concentration can be much higher than 100 nL L^{-1}.

Moreover, the ethylene concentration at the flower distributor was similar to that in ambient, which was below 10 nL L^{-1}. This provides a safe environment for the products. As Shahri & Tahir (2011) reported that flowers are not sensitivity to <10 nL L^{-1} ethylene concentration.

4.2 Broccoli responses to exogenous ethylene

Exogenous ethylene plays a significant role in influencing the postharvest quality of broccoli. Ethylene can cause chlorophyll degradation and hence the yellowing of florets (Able et al., 2005; Cai et al., 2019). Yellowing occurred the fastest and florets abscission occurred at 1000 nL L^{-1} ethylene treatment. The visual colour changes could only be seen at > 500 nL L^{-1} ethylene treatments after 14 days of exposure. An influence of 100 nL L^{-1} exogenous ethylene could be detected on colour (hue angle) after 12 days storage time measured instrumentally.

The weight loss increased with increasing ethylene concentration (Fig. 4.1). The weight loss of broccoli exposed to 1000 nL L^{-1} ethylene was 1.14 g/day, while those stored in the air (i.e. 0 nL L^{-1} ethylene) was 0.51 g/day. This is because ethylene can speed up the senescence and florescence abscission of broccoli (Li et al., 2017). Detachment of the florets from the main plant could be the reason of the high weight loss observed in high ethylene environments. In addition, Tian et al. (1995) found that large florets had a higher weight loss compared to small florets. This might also contribute to the differences in weight loss observed in our study.

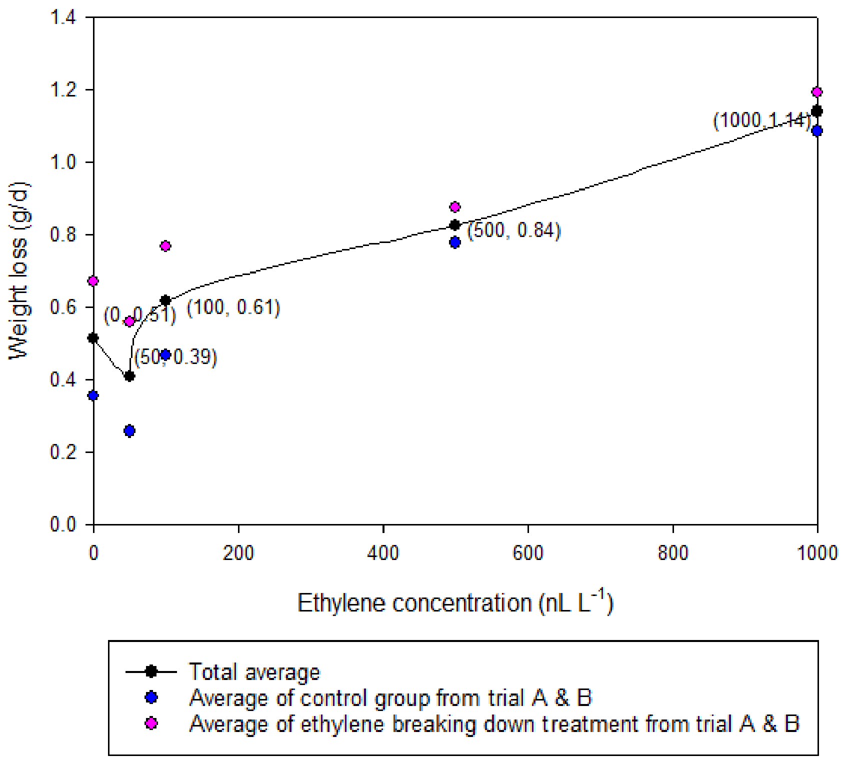


Figure 4.1 Ethylene concentration response of weight loss

Higher ethylene concentration resulted in higher rate of decrease in hue angle (h value). For instance, the h value of broccoli decreased by 5.06° per day when stored at 1000 nL L^{-1}, while that decreased 0.52° per day when stored at 0 nL L^{-1}.

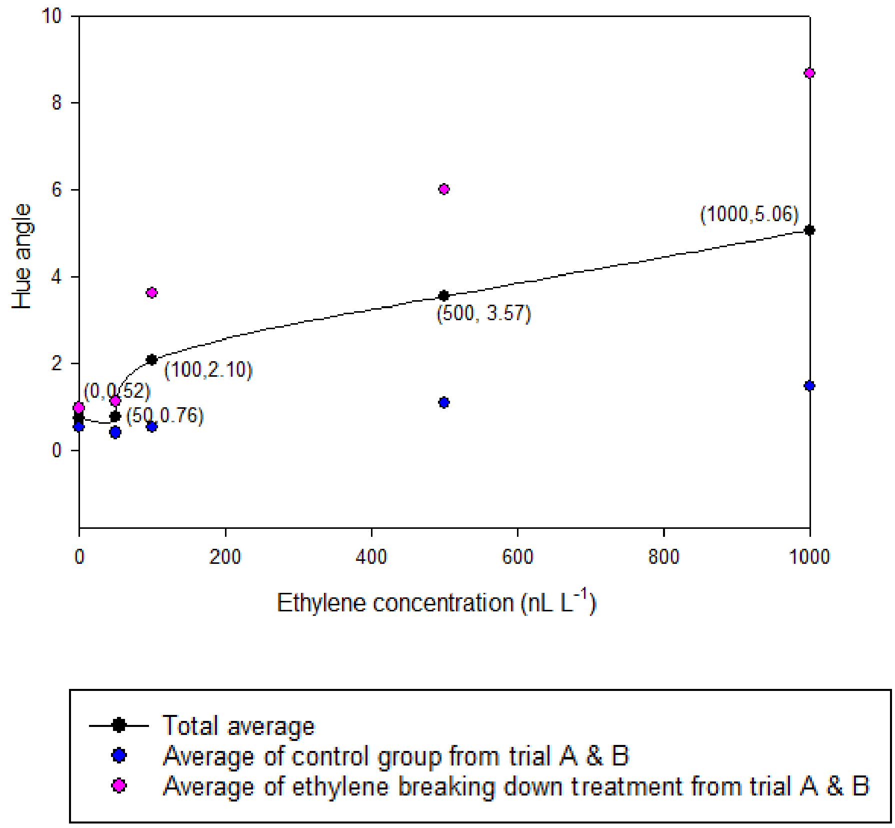


Figure 4.2 Ethylene concentration response of the decrease rate of hue angle

4.3 Ethylene effects on broccoli in the industrial supply chain

There were no significant influences on broccoli quality when the produce was stored at 0 and 50 nL L^{-1} respectively. The broccoli was sensitive to ethylene concentration ≥ 100 nL L^{-1}. This result is similar to Rees et al. (2011)'s study, as they found that the broccoli was sensitive to ethylene concentration which was around 100 nL L^{-1}.

4.4 Ethylene control in the supply chain

There are several methods to control ethylene concentration in the industrial environment. Replacing combustion engines with vents, hand or electric forklift were recommend by Blanke (2014). Wills et al. (2000) reported that vents can be an effective method to prevent products from accumulating ethylene. Besides, the application of non-toxic chemical, such as 1-MCP and AVG, can block ethylene receptors and reduce the effects of ethylene (Klee, 2005; Blanke, 2014). The use of ethylene scrubber is also used to reduce ethylene (Álvarez-Hernández et al., 2019). Blanke (2014) found that the ethylene concentration in cold store with apple reduced from 4500 nL L^{-1} to 11 nL L^{-1} with the application of ethylene scrubber.

4.5 Recommendation

Further study should investigate the ethylene responses of broccoli stored in more realistic supply chain environments. For example, the broccoli should be first stored in an environment simulating the distribution centre, followed by a short period of ambient warming simulating local transportation to supermarkets, and then stored in a temperature-controlled room resembling the chiller room conditions in supermarkets. In addition, the ethylene concentration in the produce display area in the supermarkets should be investigated to provide a more complete picture of ethylene as a contaminant in the entire supply chain. . Furthermore, the ability of application of ethylene scrubbers to reduce ethylene concentration in the supply chain should be measured.

In our study, mesh bags were used to store the broccoli. For future researchers, plastic bags could be more helpful to hold the products. This is because mesh bags can cause damage to the head of broccoli and the florets fell off as a result, potentially affecting the accuracy of the weight loss trial.

Moreover, the quality measurements were conducted in a different chiller room, as the storage room with the ethylene flow through system did not have enough space. There was a temperature change when moving the broccoli to the measurement area. For future study, quality measurement and photography should be conducted in the same room as the experimental system.

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Appendices A

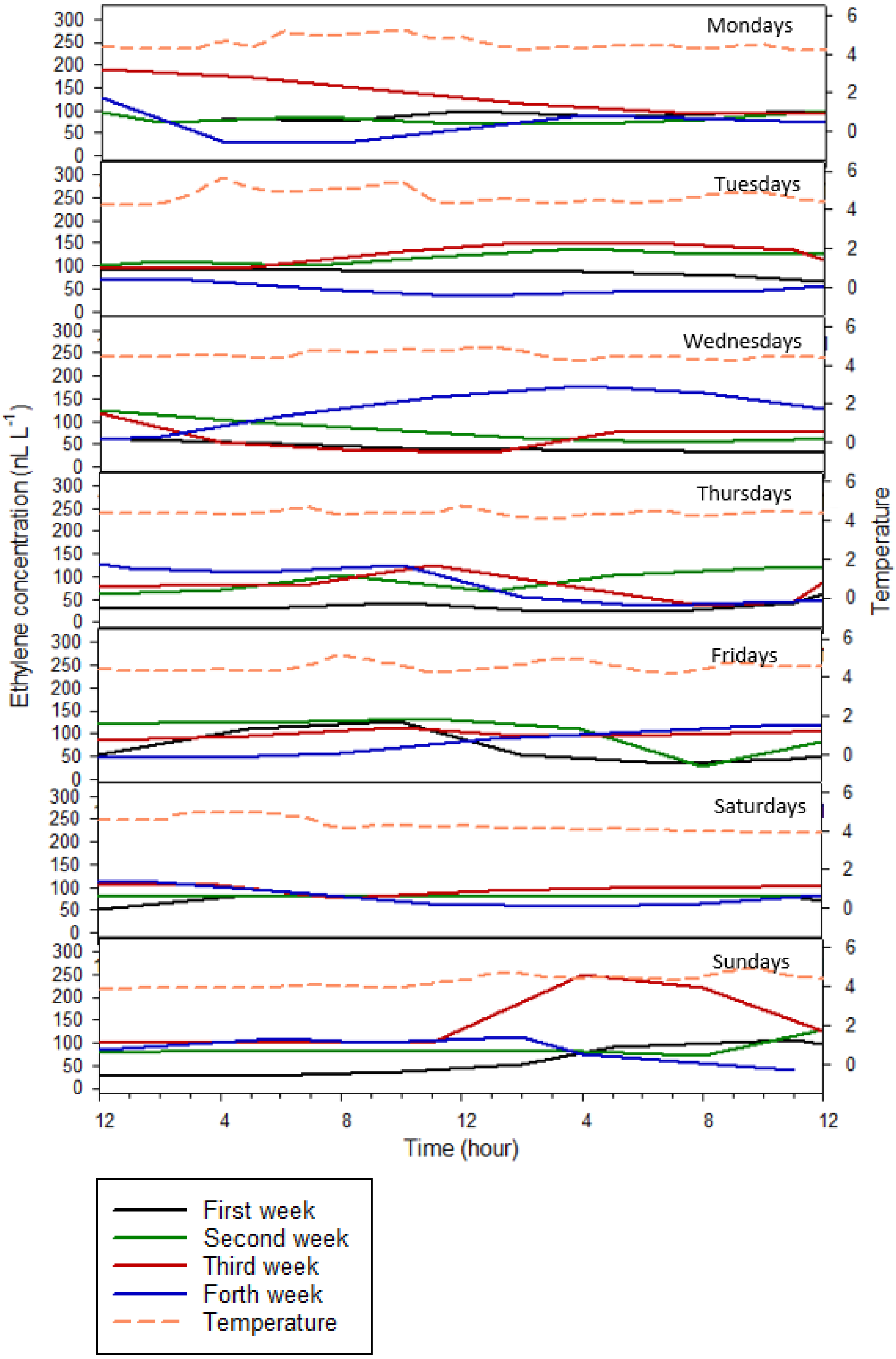

Figure A.1a The daily trend of average ethylene concentration and the average temperature of chiller room 2 (greens) in distribution centre. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 4 weeks from 13th May to 10th June.

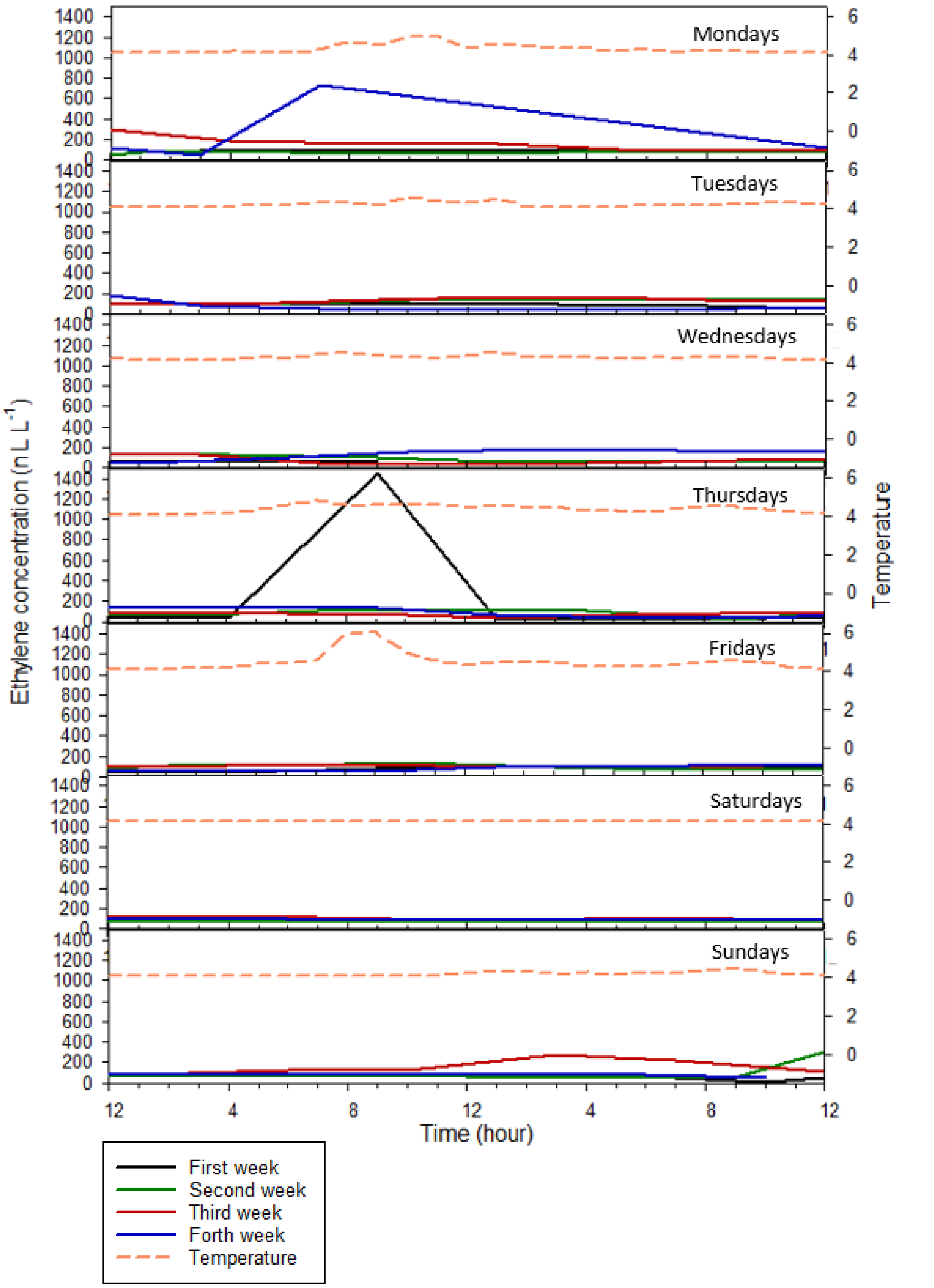

Figure A.1b The daily trend of average ethylene concentration and the average temperature of chiller room 3 (root crops) in distribution centre. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 4 weeks from 13th May to 10th June.

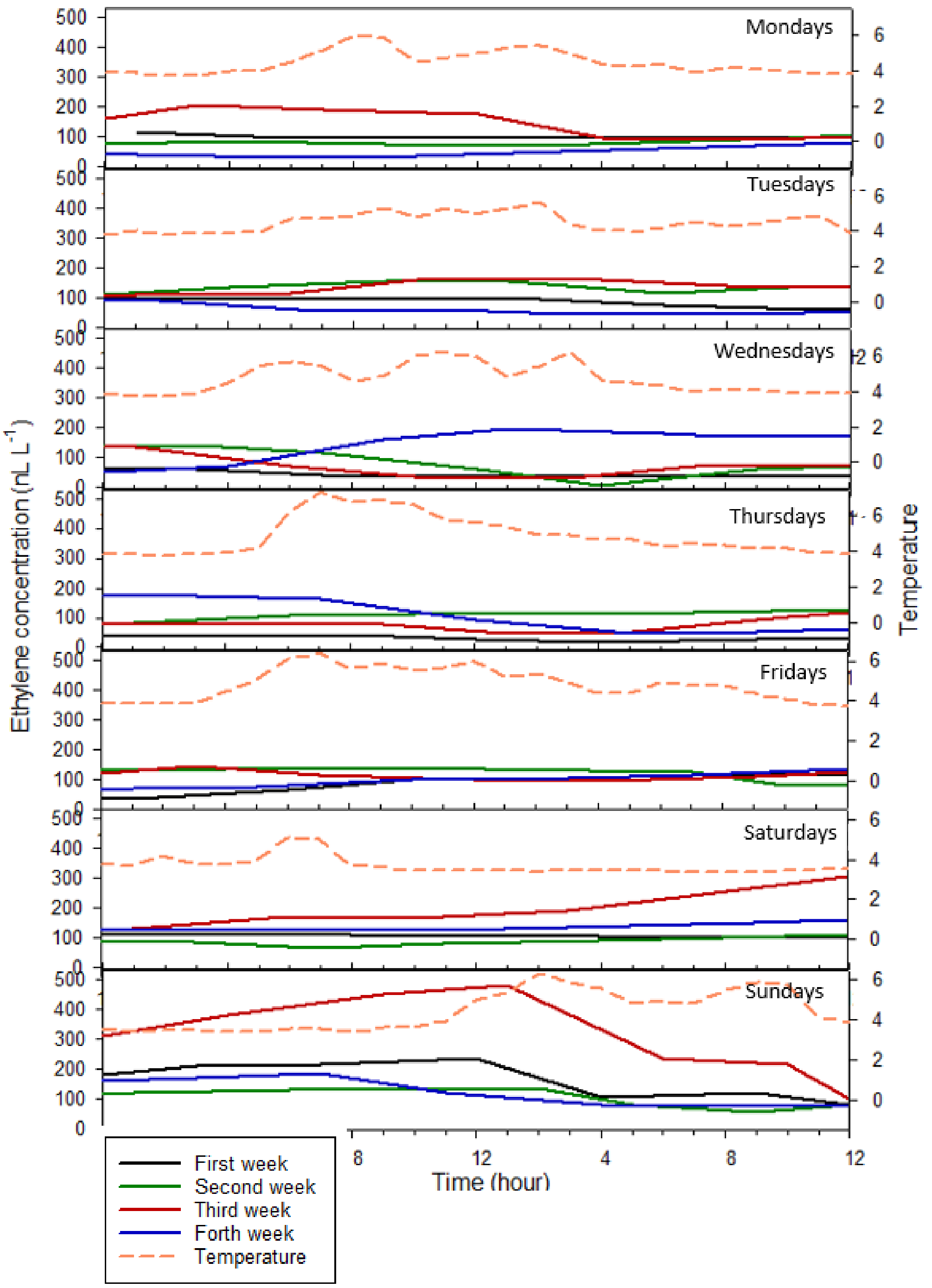

Figure A.1c The daily trend of average ethylene concentration and the average temperature of chiller room 4 (mushroom) in distribution centre. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 4 weeks from 13th May to 10th June.

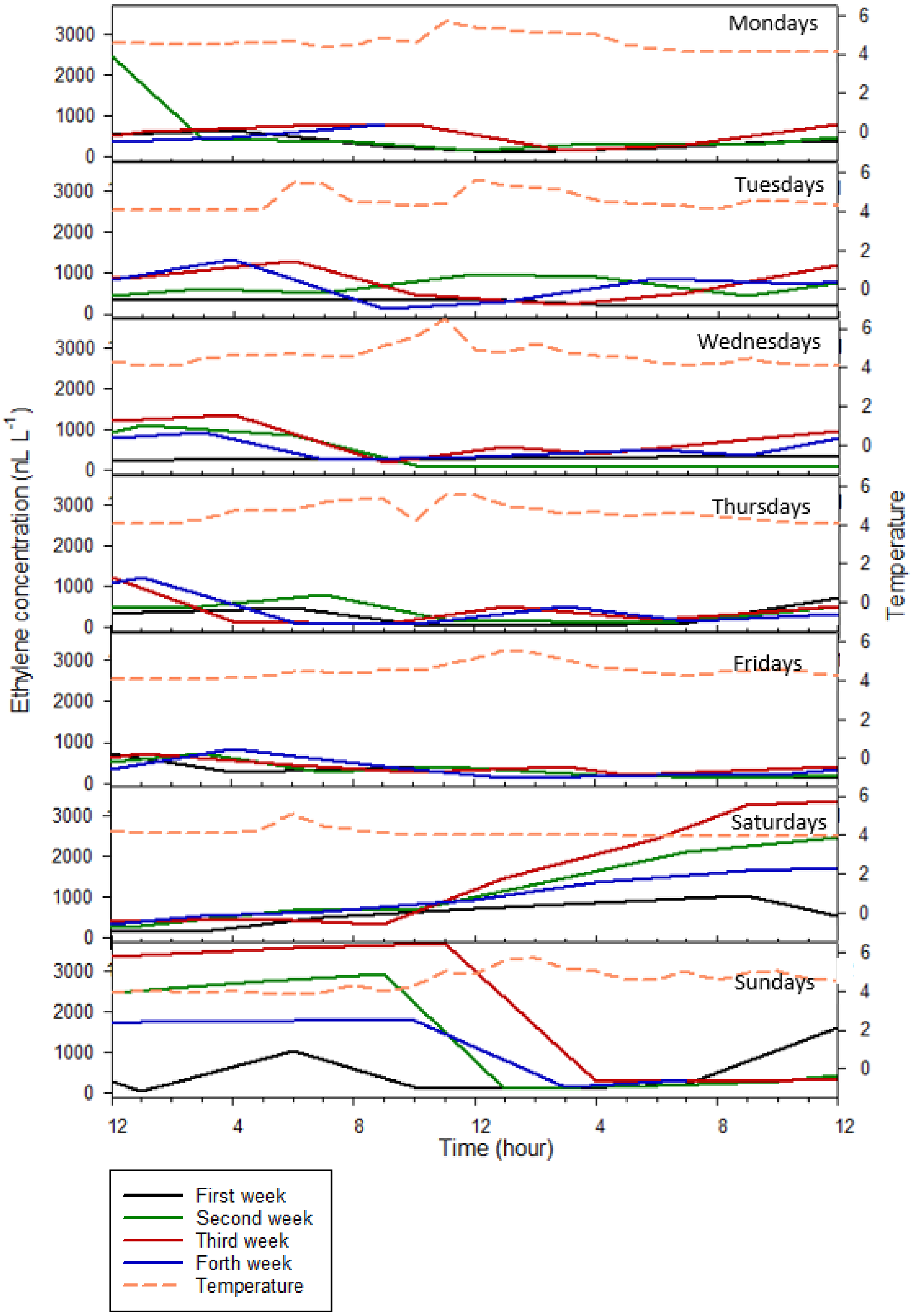

Figure A.1d The daily trend of average ethylene concentration and the average temperature of chiller room 6 (apples) in distribution centre. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 4 weeks from 13th May to 10th June.

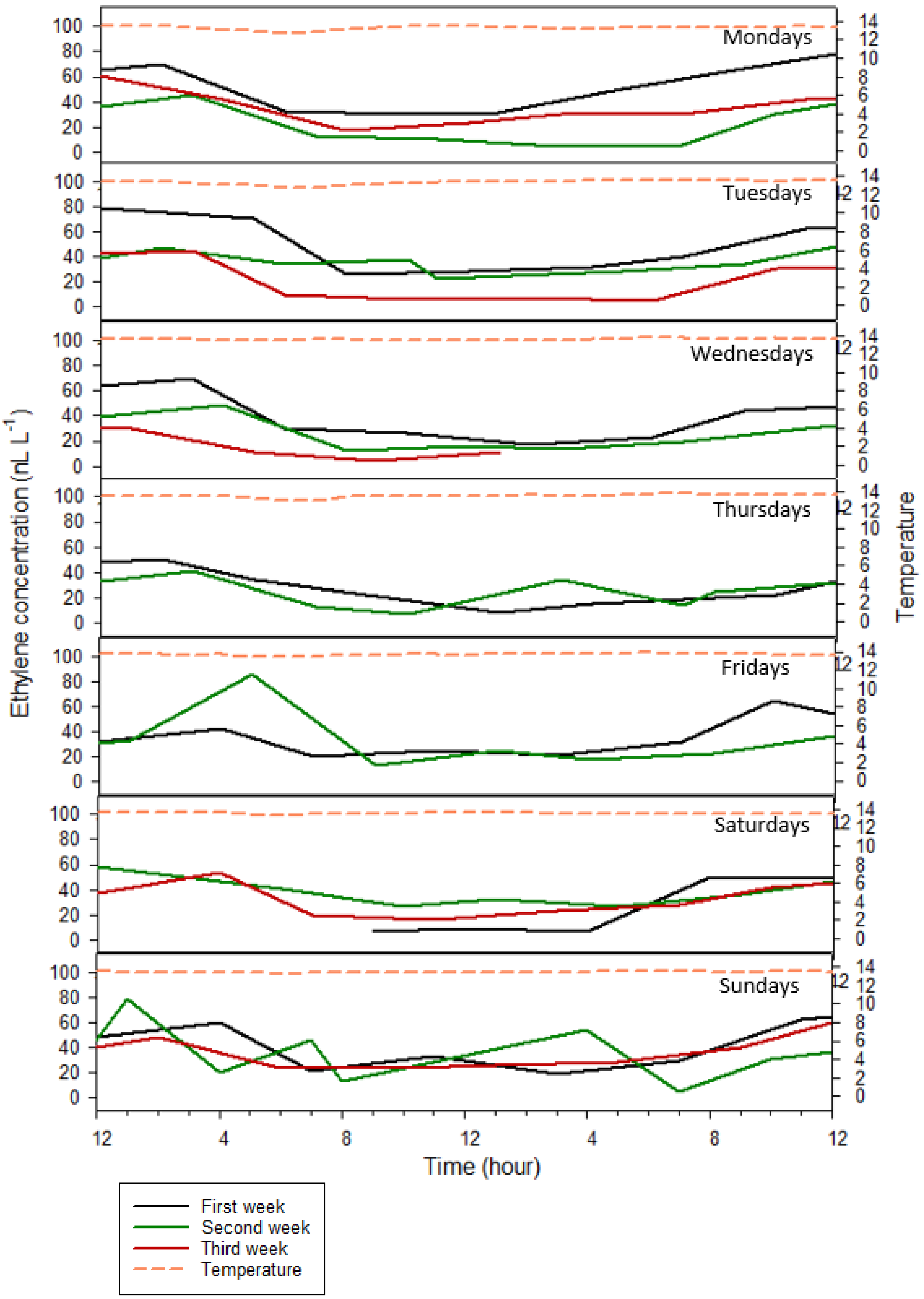

Figure A.2 The daily trend of average ethylene concentration of storage area in supermarket A. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 3 weeks from 17th July to 6th August.

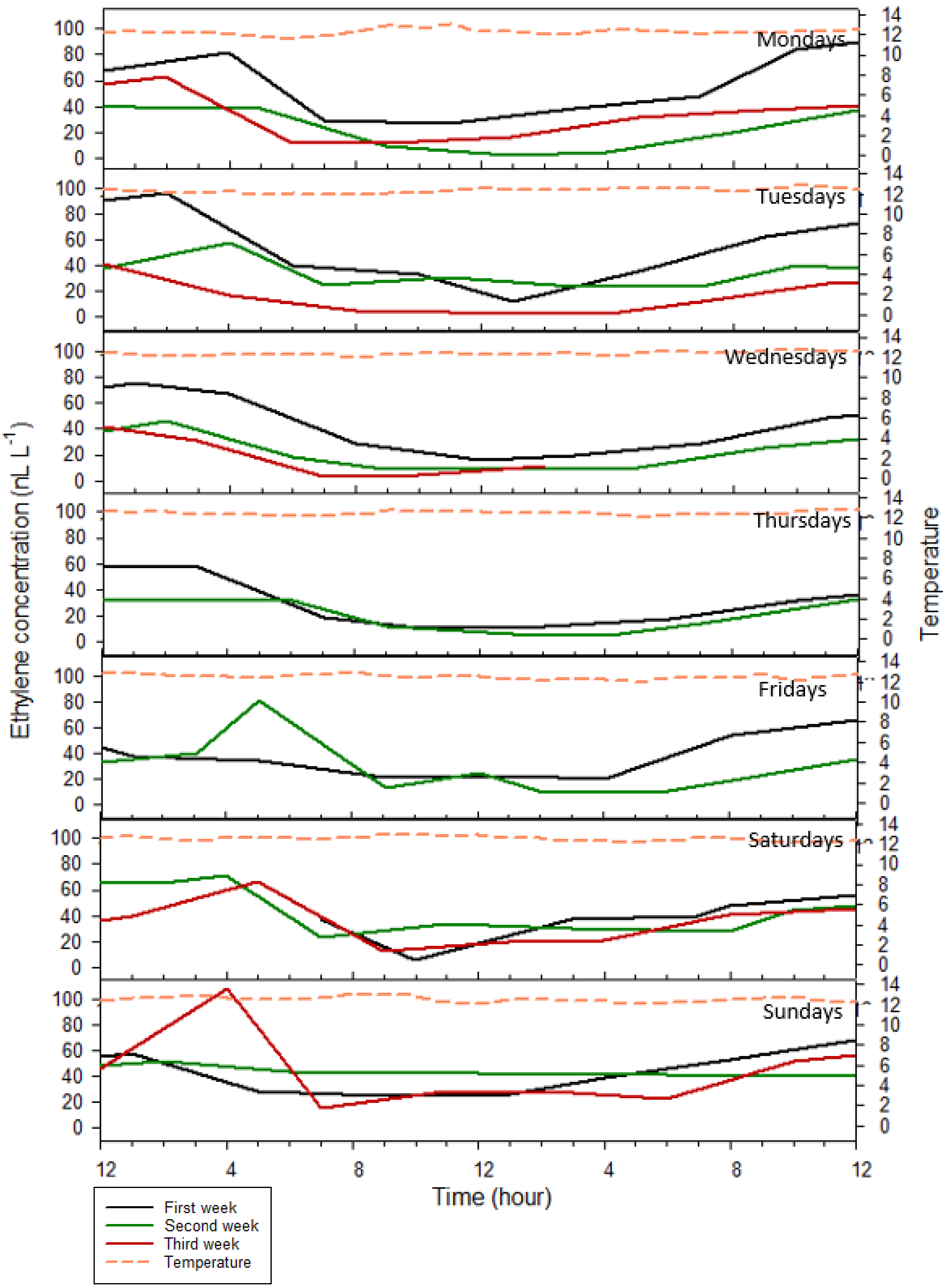

Figure A.3 The daily trend of average ethylene concentration and the average temperature of banana area in supermarket B. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 3 weeks from 7th August to 4th September.

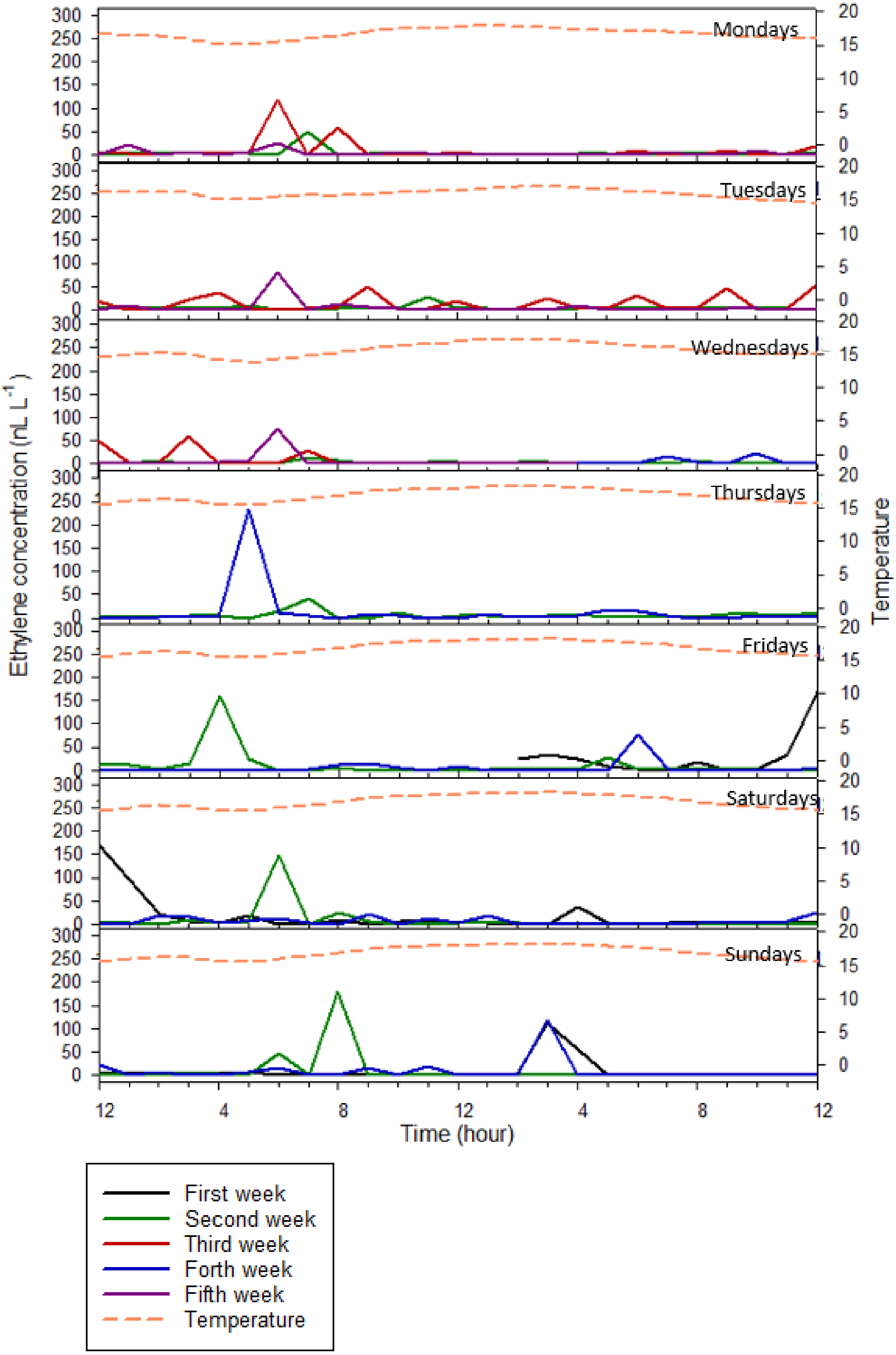

Figure A.4 The daily trend of average ethylene concentration and the average temperature of storage area in supermarket C. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 5 weeks from8th November to 4rd December.

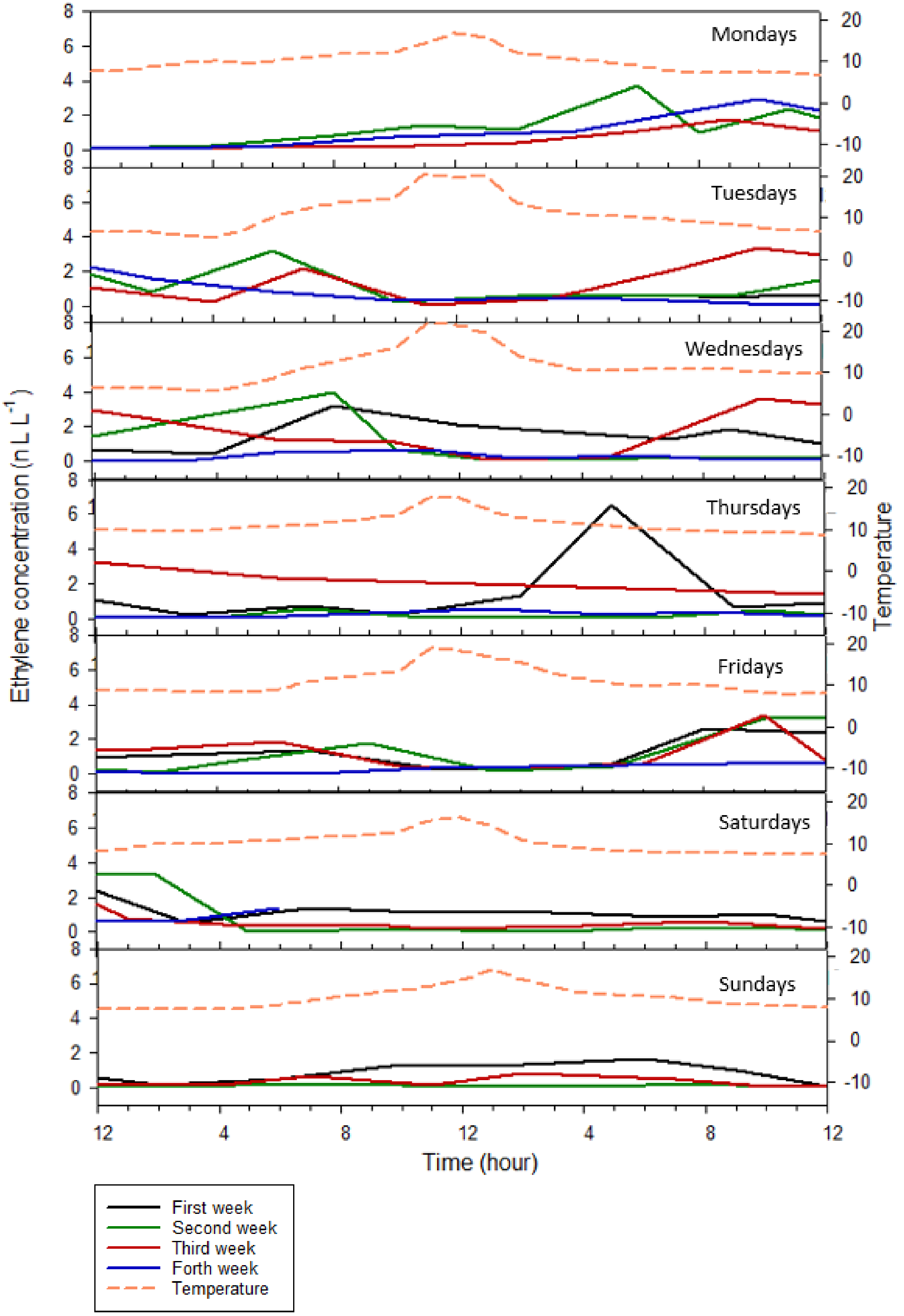

Figure A.5a The daily trend of ethylene concentration and the average temperature of ambient area in flower distributor. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 4 weeks from 3rd September to 28th September

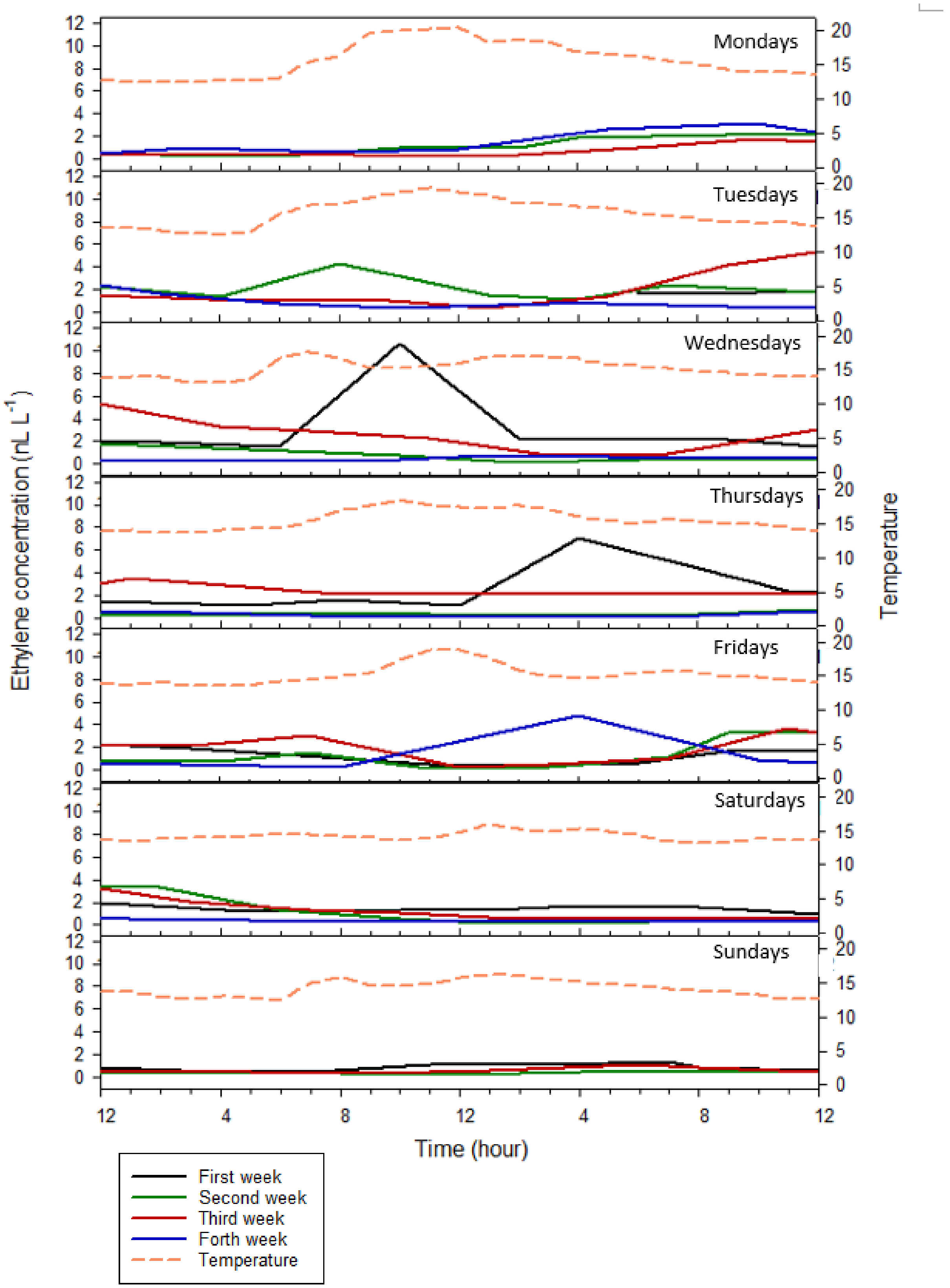

Figure A.5b The daily trend of ethylene concentration and the average temperature of preparation area in flower distributor. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 4 weeks from 3rd September to 28th September

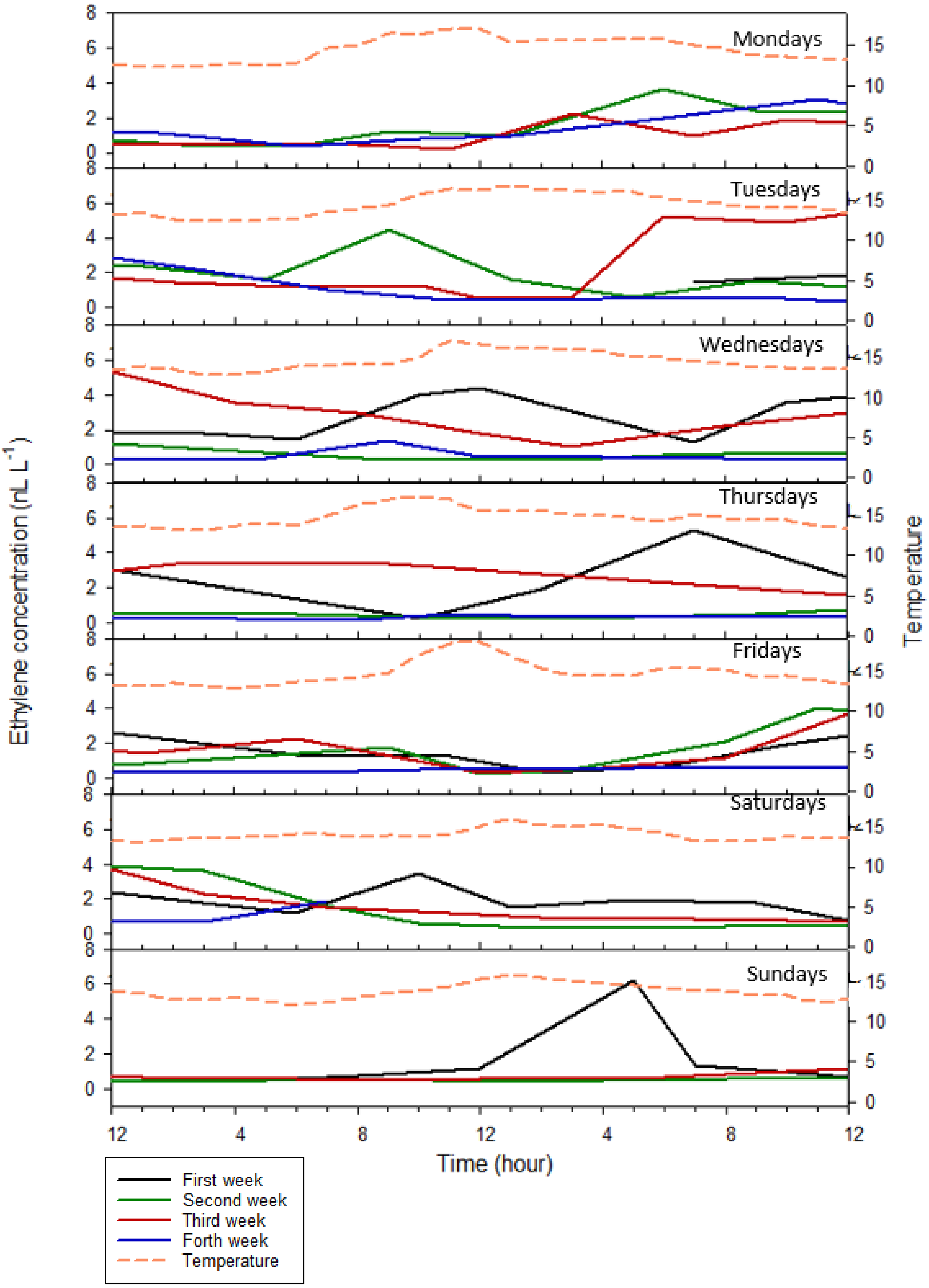

Figure A.5c The daily trend of ethylene concentration and the average temperature of display area in flower distributor. Each data point is an average of 3 readings from MACView, and the MACView took 15 mins for 1 reading. Entire data last 4 weeks from 3rd September to 28th September.

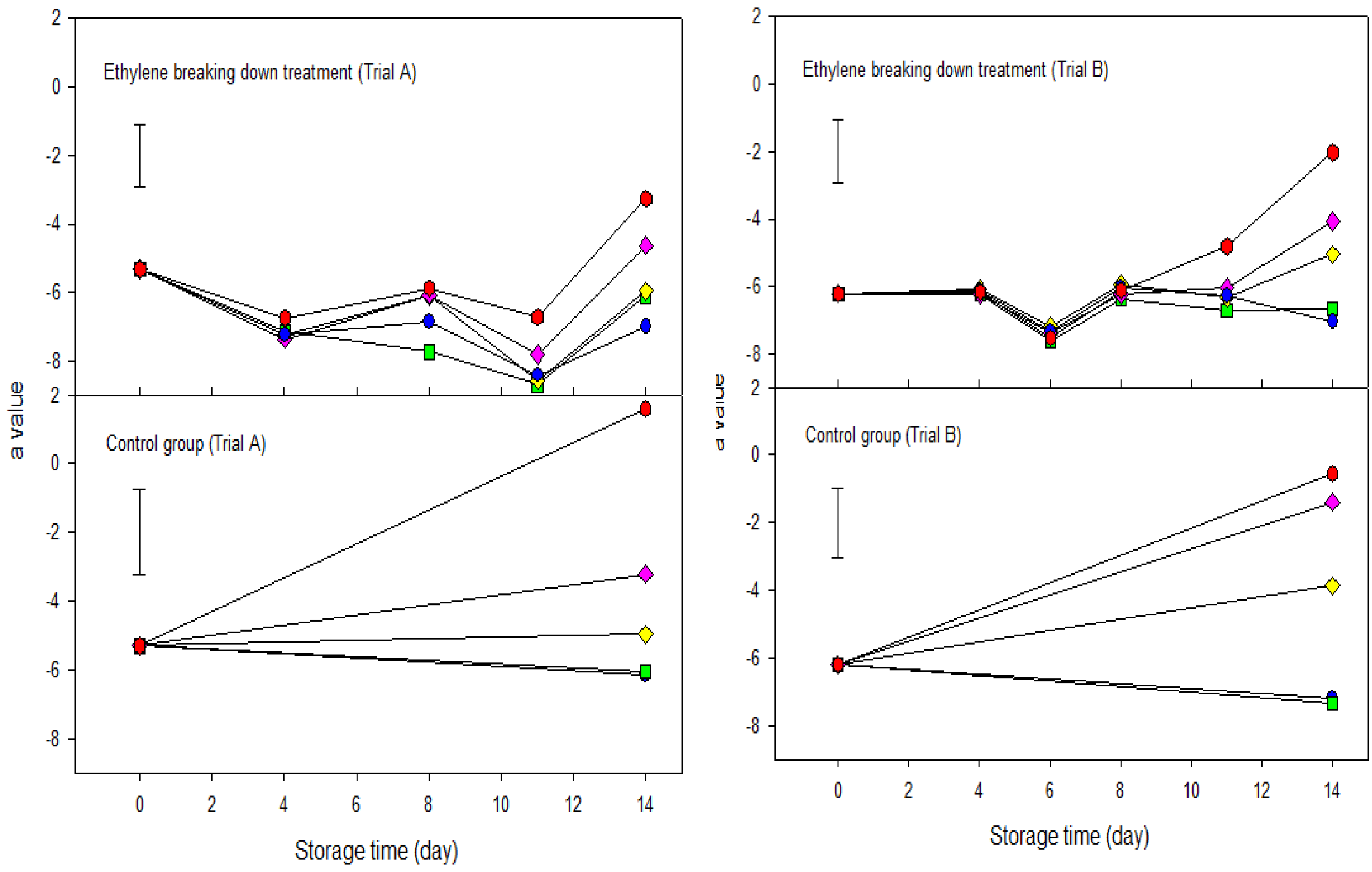

Figure A.6a a value during storage days of broccolis ethylene breaking down treatment and control group in trial 1 and Trial B at 4℃ and 98% RH. (a-b) repeated measures groups(As there was a calibration error of the colour measurements on day 6 of repeat measures group in trial A, the data was deleted from the graphs.). (c-d) single measures group. Data points are averages of 30 broccoli. $LSD_{0.05}$ has been shown as error bars.

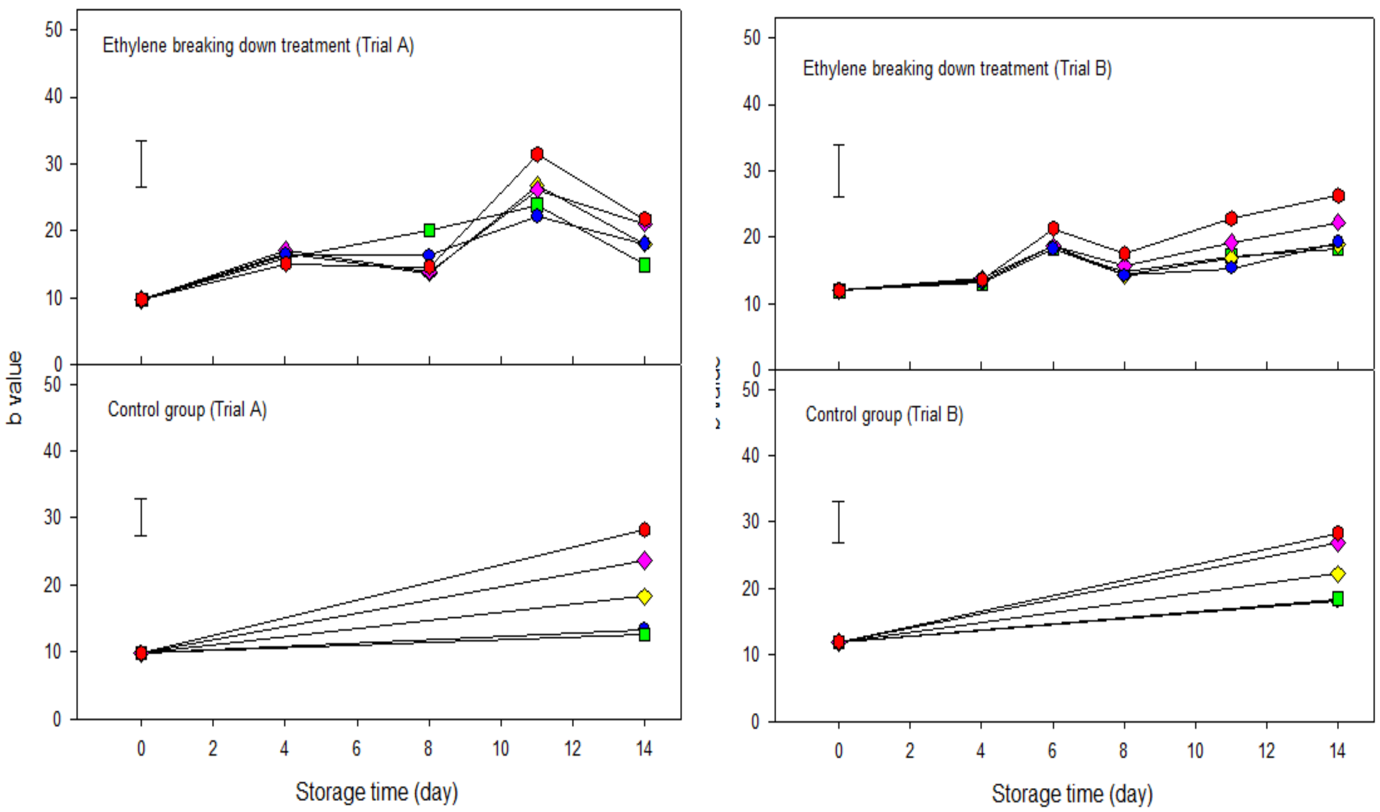

Figure A.6b b value during storage days of broccolis ethylene breaking down treatment and control group in trial 1 and Trial B at 4℃ and 98% RH. (a-b) repeated measures groups(As there was a calibration error of the colour measurements on day 6 of repeat measures group in trial A, the data was deleted from the graphs.). (c-d) single measures group. Data points are averages of 30 broccoli. $LSD_{0.05}$ has been shown as error bars.