

Review Paper

Senescence and Postharvest Studies of Cut Flowers: A Critical Review

Pooja Rani and Narender Singh*

Department of Botany, Kurukshetra University, Kurukshetra, 136119 Haryana, India

ABSTRACT

Flower senescence is the terminal phase of developmental processes that leads to the end of its life span. Since a number of developing countries are attracted to this global fresh flower trade for commercial purpose, this phenomenon is major obstacle for all the floricultural industries. Therefore, research related to postharvest changes was carried out to mitigate this problem. The post-harvest events in floricultural crops reflecting petal senescence are being reviewed in this paper, whereby various physiological and biochemical studies having data regarding lipid peroxidation, loss of membrane integrity and protein degradation central to petal senescence are included. Ultrastructural changes involving change in various cell organelles viz. rupturing of vacuole, tonoplast membrane invagination, chloroplast degradation in mesophyll cells, as well as change in mitochondria ultrastructure have also been recited. This review also pays attention to the issues related to carbohydrate metabolism and change in anthocyanin pigmentation during postharvest life. Various enzymatic activities, supporting petal senescence and current status of post-harvest technology applied to cut flowers to enhance their vase life especially by using preservatives in the form of energy source like sucrose and other sugars, biocides, mineral ions, growth regulators or various metabolic inhibitors, providing practice solution to global cut flower market, are cited.

Keywords: Cut flowers, postharvest, ultrastructural changes, senescence, biochemical changes, growth regulators

ARTICLE INFO

Article history:

Received: 4 April 2013

Accepted: 10 September 2013

E-mail addresses:

poojabishan@gmail.com (Pooja Rani),

nsheorankuk@yahoo.com (Narender Singh)

* Corresponding author

INTRODUCTION

Owing to a steady increase in demand of flowers, floriculture has become one of the important commercial trades in agriculture. Floriculture is now seen as a high growth

industry from export angle and is therefore a lucrative business. The production and export of floriculture crops from developing countries provide trade and currency. Investments in floriculture in developing countries can serve to decrease many social ills including poverty, terrorism, and illegal drug trafficking. Global exports over the last few years have grown by more than 10% annually, and at this growth rate, the world exports are expected to reach US\$ 25 billion by 2012. In order to meet this growing and changing demand, production has continued to move from countries that have traditionally been consumers and growers, such as the Netherlands, to other relatively new producing countries such as Israel, Colombia, Ecuador, Kenya, and Ethiopia. The research provides valuable knowledge about the execution of senescence in plants or plant parts like leaf, petal or sepal and how senescence is influenced by biotic and abiotic factors like environmental stresses and what physiological and biochemical changes occur during this process. This information will be used to increase the shelf life of flowering plants, which will reduce postproduction shrink and increase the profitability of floriculture producers. Therefore, since from past few years, postharvest physiology of flower has been gaining much attention to study this phenomenon of senescence and various techniques are being designed to slow down this process.

Flowers play a vital role in angiosperm reproduction; they are often pigmented and or perfumed to attract pollinators. However,

despite its irreplaceable ecological role, the flowers are energetically expensive to maintain beyond their useful life, and therefore have a limited life-span that is usually taken away after pollination; causing senescence syndrome. Senescence of flower is a complex process, so often researchers mainly concentrate on changes occurring during petal senescence. Petals provide an excellent model system for the study of fundamental aspects of senescence (Rogers, 2006; Desai *et al.*, 2012). Senescence is a highly regulated final event of flower development that bears hallmarks of programmed cell death (PCD), resulting in colour changes, petal wilting, abscission of whole flower and flower parts with various physiological, biochemical and ultrastructural changes (Voleti *et al.*, 2000; Wagstaff *et al.*, 2003; Jones *et al.*, 2005; Tripathi & Tuteja, 2007; Seo *et al.*, 2009; Ichimura, 2010; Shahri, 2011). Recent studies evidenced that flower senescence includes controlled disassembly of cells of corolla probably by a mechanism homologous with apoptosis, vacuolar and necrotic PCD (Van Doorn, 2011), and transport of nutrient to other parts of inflorescence. The most important barriers in the marketing and commercialization of many cut flowers are their short vase life and their inability to withstand stresses during storage or transit (Halevy & Mayak, 1981; Nowak & Rudnicki, 1990; Zamani *et al.*, 2011). A great deal of research dealing with best post-harvest care of cut flowers has been carried out in recent years but our understanding of cut flower physiology is

still quite rudimentary, despite development of techniques that have enabled us to maintain good cut flower quality, longer than ever before. Petals are the main floral organs which primarily determine the commercial longevity of flowers and as a consequence, it becomes necessary to study the physiological, biochemical and genetic processes that occur during petal senescence (Chakrabarty *et al.*, 2009) and how it can be slowed down through designing inexpensive postharvest technologies that ultimately extend the postharvest life of cut flowers (Wani *et al.*, 2012). This review is expected to give an update of literature on postharvest behaviour of cut flowers and some of the recent technologies contributing to their postharvest life which is major priority for growth of such a global floriculture industry. It also comprehends metabolic changes regarding protein degradation, lipid peroxidation, alteration in sugar levels in phloem exudates, activity of various enzymes and colour change central to petal senescence during post-harvest life of cut flowers to know exact the mechanism of senescence during post-harvest life and also entails the use of various preservatives or holding solutions like sugars, biocides, mineral ions, growth hormones and metabolic inhibitors, etc. to mitigate the problem of short post-harvest life and how they serve to retard the petal senescence. There are some other practices are not discussed here.

POST-HARVEST CHANGES ASSOCIATED WITH SENESCENCE

Ultrastructural Changes

There is a three-stage theory of senescence in case of flowers like those in leaf. First is the initiation of senescence followed by degradation and disassembly which lead to third stage of death (Yoshida, 2003), which is due to decline in rate of anabolic processes and increase in rate of certain catabolic processes (Galston & Davies, 1970). Characteristics of the last phase involve ultrastructural disorganization of tissues or cells and increased fluid filled extra spaces which lead to halted down of all metabolic activities in all tissues or organs of plant. But, some organelles are still slightly visible (Smith *et al.*, 1992). Delicacy of petal cells and their rapid collapse during senescence are a challenge to study ultrastructural changes during senescence and showed dramatic changes in ultrastructure (Van Doorn *et al.*, 2003). Ultrastructural events during senescence include increase in vacuolar size, loss of organelles, eventual collapse of tonoplast (Van Doorn & Woltering, 2004) and nuclear fragmentation (Yao *et al.*, 2004; Yamada *et al.*, 2006; Battelli *et al.*, 2011). Wiemken *et al.* (1976) used *Iris* as a model system to study ultrastructural, biochemical and molecular changes during senescence. One of the earliest changes in ultrastructure of *Iris* petals is closure of plasmodesmata. Plasmodesmata if open allow transfer of small molecules like sugars, hormones and RNA molecules between adjacent cells. If plasmodesmata are closed, transport

gets halted. Ultrastructural work in *Iris*, *Sandersonia*, senescing corolla of *Lycoris longituba* Y.C. Hsu & G.J. Fan and *Lilium longiflorum* also showed complete degradation of wall of mesophyll cells prior to visible senescence due to closure of plasmodesmata whereas epidermal cells remain intact (Wagstaff *et al.*, 2003; Van Doorn *et al.*, 2003, O'Donoghue *et al.*, 2005; Lei *et al.*, 2009; Battelli *et al.*, 2011). Other ultrastructural events involve invagination in tonoplast and presence of numerous vesicles in vacuole which is main site of organelle degradation (Matile & Winkenbach, 1971; Smith *et al.*, 1992). As senescence proceeds, all cytoplasmic content get lost in carnation (Smith *et al.*, 1992) and *Iris* (Van Doorn *et al.*, 2003). The increases in the number of small vacuoles and size of vacuole have also been evidenced in carnation (Smith *et al.*, 1992), *Iris* (Van Doorn *et al.*, 2003) and *Hemerocallis* (Stead & Van Doorn, 1994). Meanwhile, disappearance of free ribosomes and clusters attached to endoplasmic reticulum during maturation and senescence followed by reduction in the number of mitochondria, golgi bodies followed by other organelles, was also recited (Butler & Simon, 1971; Smith *et al.*, 1992; Van Doorn *et al.*, 2003). Most noticeable changes during development and senescence take place in plastids show invaginations in plastid membrane. Changes in the structure of tissue containing chromoplasts have also been observed in cucumber petals (Smith & Butler, 1971) and amyloplasts in *Lycoris longituba* Y.C. Hsu & G.J. Fan (Lei *et al.*,

2009). With reference to the chloroplast ultrastructure, a higher level of thylakoid disorganization (especially of granal membranes) is observed during senescence (Spundova *et al.*, 2003). The loss of non-cellulosic natural sugar and the increase in soluble pectin, uronic acid and cellulose (De Vetten & Huber, 1990; Smith *et al.*, 1992) lead to alternation in cell wall and initiation and increase in the loss of membrane integrity resulting in phase changes due to decrease in membrane fluidity and increased permeability (Thompson *et al.*, 1982; Knowles *et al.*, 2001). The event related to the loss of RNA and DNA present in nucleus, mitochondria, chloroplast and plastids is also associated with senescence (Rubinstein, 2000; Aleksandrushkina *et al.*, 2008; Lei *et al.*, 2009) before or after vacuolar collapse. More detailed analysis should be carried out at cell and organelle levels to determine exactly what is happening inside the petal during senescence and how we can retard this syndrome of senescence.

Colour Fading and Change in Pigmentation

Chlorophyll is the most prominent photosynthetic pigment in higher plants and the decreasing trend in photosynthetic rate and pigment level is generally due to the involvement of oxygen radicals and singlet oxygen (Prochazkova *et al.*, 2001; Dertinger *et al.*, 2003). Chlorophyll breakdown becomes a mandatory phenomenon for the remobilization of nitrogen from chlorophyll-binding proteins to proceed during senescence (Hortensteiner, 2006). Louda *et al.* (2002) and Spundova *et al.*

(2003) found that most chlorophyll species were broken down during senescence. However, considerable delay in degradation of chlorophyll compared with control and copper sulphate was found by keeping the flower in aluminium sulphate and ethanol by slowing down the rate of transpiration and deterioration (Hajizadeh *et al.*, 2012).

Colour fading and discoloration are major reasons for termination of vase life in many cut flowers and important factors in determining display quality of cut flowers. Major three types of pigments contributing to the colour of flowers are anthocyanins, carotenoids, and betalains. Anthocyanins are the largest and most diverse group of plant pigments derived from the phenylpropanoid pathway, ranging in colour from red to violet and blue. Anthocyanins give red colour under low pH and blue colour under high and neutral pH, reflecting a phenomenon termed as 'blueing' where a shift from red to blue occurs with ageing (Wills *et al.*, 1998; Avila-Rostant *et al.*, 2010). Ultimate flower colour of a species is determined not only by the pigment present but also by various other factors like pH (Harborne, 1988; Yoshida *et al.*, 2003; Katsumoto *et al.*, 2007), temperature (Shvarts *et al.*, 1997; Dela *et al.*, 2003), light (Weiss, 2000; Meng & Wang, 2004; Irani & Grotewold, 2005), and mineral ions (Shoji *et al.*, 2007). Griesbach (2005) observed that although flavonols and an appropriate pH are important in obtaining blue orchids, the more important of the two factors was vacuolar pH. In morning glory, the colours of flower vary from reddish purple in buds to blue in flowers with an increase in vacuolar pH from 6.6

to 7.7, a change believed to be driven by a $\text{Na}^+(\text{K}^+)/\text{H}^+$ exchanger (Yamaguchi *et al.*, 2001; Yoshida *et al.*, 2005). Collette *et al.* (2004) and Elibox and Umaharan (2008) investigated the relationship between epidermal vacuolar pH and a number of plant factors in anthurium with the intention of creating colours in the blue range by characterizing the genetics and biochemistry of the anthurium flavonoid biosynthetic pathway. In flowers, degradation of anthocyanins during senescence is possibly related to oxidative process. A significant increase in antioxidant activity is correlated with the rate of anthocyanin degradation (Mazza, 2007). Reducing agents such as glutathione can inhibit the degradation of anthocyanins (Vaknin *et al.*, 2005). In morning glory (*Ipomoea tricolor*) petals, the vacuolar pH is relatively low when the flower bud opens, resulting in a red colour but upon further maturation, the vacuolar pH increases and the petals acquire a strong blue colour, but in mutants, colour change does not occur and remains purple (Yoshida *et al.*, 1995).

Rose flowers produced under cooler environment have higher anthocyanin content especially during summer (Plaut *et al.*, 1979). High temperature applied at different stages of flower development reduces anthocyanin content in petals (Dela *et al.*, 2003). High temperature and low light conditions have also been reported to reduce pigment content in petals. This is due to the breakdown and down regulation of genes encoding enzyme involved in biosynthesis of anthocyanins (Gonzalez, 2009).

Low light intensity plants develop pale flowers with a low level of anthocyanins (Biran & Halevy, 1974; Griesbach, 1992). In *Eustoma grandiflorum* Shinn., low light conditions result in reduced anthocyanin content in petals of developing florets both in cut flowers and potted plants measured by the expression pattern of six genes encoding enzymes for anthocyanin biosynthetic pathway in developing petals concluded that light intensity regulates a master transcription factor common for all these anthocyanin biosynthesis genes (Meir *et al.*, 2009). Low light intensity affects petal pigmentation through reduced photosynthesis in the leaves or stems, which in turn reduces the soluble sugar content of petals and leads to a repression of the genes that encode enzymes of anthocyanin biosynthetic pathway in *Eustoma grandiflorum* (Kawabata *et al.*, 1995). Therefore, increased sucrose concentration has been found to enhance petal growth and pigmentation in detached flowers of *Eustoma grandiflorum* and rose (Kuiper *et al.*, 1991; Sankhla *et al.*, 2005). The influence of sucrose and light intensity on lightness, chroma and petal colour of flower has also been reported through change in anthocyanin pigmentation in lilianthus cultivars (Uddin *et al.*, 2001). The induction of anthocyanin synthesis and anthocyanin biosynthetic gene expression in detached petunia (*Petunia hybrida*) corollas by gibberellic acid (GA₃) requires sucrose for activation of anthocyanin biosynthetic gene (Moalum-Beno *et al.*, 1997).

Carotenoids like anthocyanins are also widely distributed in angiosperms

whereas betalains are only found in some plants in the order Caryophyllales in some higher fungi such as *Amanita muscaria*. Betalains have functions analogous to those of anthocyanins as pigments. The majority of carotenoids in the petals of sandersonia (*Sandersonia aurantiaca*) are β , β -carotenoids such as β -cryptoxanthin, zeaxanthin and β -carotene (Nielsen *et al.*, 2003). On the other hand, more than 90% of the carotenoids in the petals of (*Tagetes* sp.) marigold (Moehs *et al.*, 2001) and chrysanthemum (Kishimoto *et al.*, 2004) are lutein and/ or lutein derivatives. There are many carotenoids whose biosynthesis has not been characterized.

Taken together, it is concluded that anthocyanins, carotenoids and betalains constitute the majority of the flower pigments which can be affected by various factors like pH, light, temperature, etc. Some preservatives like sugar and growth hormones like GA₃ might prove to be best in delaying degradation of pigments like anthocyanins. At present, biosynthetic pathway of these pigments and their regulation are well known. However, the mechanisms of transport and sequestration of these pigments inside the vacuole which petals show variable colour is not known. Molecular approach may prove to be beneficial to know the mechanism of transport and their sequestration.

Lipid Peroxidation and Loss of Membrane Integrity

Lipid peroxidation generates a range of reactive oxygen species, including singlet oxygen (¹O₂), the alkoxy radical (RO[•]),

hydroxyl radical (OH[•]) and hydroperoxyl radical (HO^{2•}), and the peroxy radical (ROO[•]) which amplify the lipid peroxidation with further degradation of released fatty acids affecting membrane permeability (Van Doorn & Woltering, 2008; Rogers, 2012). All these can oxidize a range of macromolecules with varying specificity, although the hydroxyl radical is the most reactive and least specific (Dat *et al.*, 2000). Bialeski and Reid (1992) found evidence for rapid cessation of overall phospholipids synthesis during petal senescence in *Hemerocallis*. All the enzymes required for phospholipid degradation are apparently present in membranes, even in those of young cells (Brown *et al.*, 1990). Microsomal membranes from the petals of senescing carnation (*Dianthus caryophyllus* L.) flowers contain phosphatidylcholine, phosphatidylethanolamine, phosphatidylglycerol and phosphatidylinositol. These phospholipid classes decline essentially together during natural senescence of the flower (Brown *et al.*, 1991; Chakrabarty *et al.*, 2009; Lei *et al.*, 2009). The decrease in levels of various lipids is associated with plant senescence. During petunia (*Petunia hybrida* Vilm.) flower senescence, there is a senescence-related increase in the content of diacylglycerol, one of the products of phospholipids metabolism in plasma membranes (Borochoy *et al.*, 1997).

The main phospholipid degrading enzymes, including (i) Phospholipase D (ii) Phospholipase C (iii) Lipolytic acyl hydrolase and (iv) Lipooxygenase, which degrade fatty acids were reported

to be upregulated during petal senescence. Degradation of free fatty acids mainly occurs by β -oxidation (Pistelli *et al.*, 1992), end product of which is fed into glyoxylate cycle and converted to sugars in cytosol (Chen *et al.*, 2000; Cornah *et al.*, 2004) or used in synthesis of amino acids. Acyl-CoA oxidase isozymes catalyse the first step in peroxisomal fatty acid β -oxidation. The transcript abundance of a gene encoding an acyl-CoA synthase increased during senescence making plasma membrane permeable prior to vacuolar collapse due to phospholipid degradation inside membrane (Hopkins *et al.*, 2007). Freeze-fracture electron microscopy of senescing carnation petals indicated the presence of gel phase lipid in plasma membrane, ER and tonoplast showing senescence syndrome (Hopkins *et al.*, 2007). The ratio of saturated/unsaturated fatty acids was also found to increase due to extensive degradation of phospholipids and galactolipids during flower senescence (Leverentz *et al.*, 2002).

Cellular membranes are selective, dynamic barriers, structural integrity of which is necessary for critical membrane functions such as maintaining the cell's osmotic balance in petals. The rupture of cellular membrane is likely to be deleterious to plant function since regulation of metabolites and signal exchange between neighbouring cells will also be lost. Membrane damage may occur early or later in the process of senescence. The accumulation of peroxidized lipids and products derived from them in senescing membranes appears to cause extensive destabilization of the membrane bilayer

structure and loss of membrane function. Thompson (1988) also observed a strong correlation between membrane leakiness and phospholipid breakdown in senescent flowers, rendering the membrane more susceptible to lipid degrading enzymes such as lipoxygenase that leads to broken down of cell membrane (Hildebrand & Hymowitz, 1982; Fobel *et al.*, 1987; Lei *et al.*, 2009). Electrical conductivity of the petal diffusates reached maximum at full bloom, with significantly higher values in *Rosa damascene* (Sood *et al.*, 2006). Membrane permeability of sepal tissues estimated as electrical conductivity of ion leachates was also observed to increase as the development proceeded through various stages in *Ranunculus asiaticus* L. and *Consolida ajacis* L. cv. 'Violet Blue' (Shahri & Tahir, 2011a, b). Fukuchi-Mizutani *et al.* (2000) noticed increased activity of lipoxygenase (LOX) with senescence of cut flowers stimulating deterioration of membrane. Deterioration of cellular membranes causes increased membrane permeability, loss of ionic gradients and decreased function of key membrane proteins (e.g., ion pumps). Nevertheless, it is pertinent to note that loss of membrane function in *Alstroemeria* has been shown to occur without increased activity of lipoxygenase suggesting that the loss of membrane integrity can be achieved in a number of ways (Leverentz *et al.*, 2000). Membrane breakdown in lupin (Hernandez-Jimenez *et al.*, 2002), carnation (Bartoli *et al.*, 1995), day lily (Rubinstein, 2000), rose (Sood *et al.*, 2006) and gladiolus

(Hossain *et al.*, 2006) is evidenced by positive role of lipoxygenase in promoting senescence. With loss of the integrity of cellular membrane structure in cells undergoing senescence, hydrolytic enzymes being normally compartmentalized in these cells are released and could cause massive breakdown of various cellular components. There are various changes related to membrane in plant tissue. In some cases, lipid pentadienyl, peroxy free radicals may also be generated by LOX action (Roy *et al.*, 1994). The result obtained by Brown *et al.* (1987) evidenced the microsomal membrane from carnation produced increased quantity of O_2^- during senescence. According to the authors, O_2^- is produced by membrane bound oxidase. Lipid peroxidation not only threatens membrane proteins and functioning and integrity of membrane but also produces a variety of toxic aldehyde and ketones (Wilhelmova *et al.*, 2006). A marked deterioration of the plasma membrane and loss of water are associated with lipid peroxidation with the senescence of chrysanthemum and *Heimerocallis* petals (Bartoli *et al.*, 1995; Bartoli *et al.*, 1997; Chakrabarty *et al.*, 2009). Therefore, membrane degradation may be a central step in the process of senescence that leads to mass lipid degradation during senescence and collapse of the tonoplast which results in executing the death sentence. The main controversy is about the main site of phospholipid degradation and to what extent this phospholipid degradation takes place inside the vacuoles and autophagosomes and to what extent inside the membrane.

Loss of Cellular Protein

Proteins are the key molecules that play important roles in various structural and functional aspects of plants. Senescence in tepals, stamens and carpels results in an increase in total protease activity and a decrease in total protein content. In many species of plants, protein degradation and remobilization are mediated through protein ubiquitination and the action of specific proteases (Wagstaff *et al.*, 2002; Pak & Van Doorn, 2005; Batelli *et al.*, 2011) transferring various amino acids to phloem. The soluble proteins registered a consistent decrease with the simultaneous increase in specific protease activity and α -amino acid content during different stages of flower development and senescence in *Ranunculus asiaticus* L. and *Consolida ajacis* cv. Violet Blue (Shahri & Tahir, 2011a, b). In many species of other flowers like *Petunia* and rose, a drastic decrease was found in protein level prior to visible senescence symptoms (Jones *et al.*, 2005; Sood *et al.*, 2006). An increase in amino acid content in phloem exudates from flower opening to petal wilting in *Ipomoea*, *Hemerocallis* and *Sandersonia* petals was observed due to protein degradation. Asparagine, lysine, glutamine and hydroxyproline were main transport amino acids (Wiemken *et al.*, 1976; Bielecki, 1995; Eason *et al.*, 2002). Several genes like DEAD/DEAH domain helicase related to protein synthesis are differentially expressed during petal senescence, both in daffodil (Hunter *et al.*, 2002) and in *Alstroemeria* (Breeze *et al.*, 2004). These genes are homology to the

eIF-4A. Actinomycin D, an inhibitor of transcription, if given 4 h prior to opening, suppressed the onset of visible senescence, which occurred at about 9 h after flower opening by downregulation of senescence associated genes in petals of morning glory (Yamada *et al.*, 2007). SDS-PAGE of protein extract from sepal tissues of *Helleborus orientalis* suggested a decrease in the expression of high molecular weight proteins and an increase in low molecular weight proteins during flower development and senescence. At present, it is not known whether the polypeptides that increased during senescence play an important role in the senescence of *Helleborus orientalis* flowers but these polypeptides may be linked to longevity (Shahri *et al.*, 2011).

In proteasomes dependent degradation, proteasome system involved in degradation of specific proteins was apparently up-regulated during petal senescence in (*Hemerocallis*) day lily (Müller *et al.*, 2004) and daffodil (Hunter *et al.*, 2002). In carnation, the abundance of mRNA increased three genes encoding subunits of the 19S regulatory particle, one of two large complexes of the 26S proteasome (Hoeberichts *et al.*, 2007). A significant delay in the time to visible senescence was observed through feeding the isolated *Iris* petals with Z-Leu-Leu-Nva-H, an inhibitor of proteasome activity (Pak & Van Doorn, 2005). Ubiquitinated proteins involved in the degradation of many petal proteins during floral development and senescence increased in intensity as the flowers senesced. Several monomers of

ubiquitin (a 76 amino acid polypeptide) become attached to protein targeted for degradation in the proteosomes having involvement of three enzymes referred to as E1, E2 and E3. Silencing of RING domain of E3 protein delayed visible senescence symptoms in *Petunia* (Xu *et al.*, 2008). Expression of a homologous gene encoding a RING zinc finger ankyrin repeat protein (MjXB3), a putative E3 ubiquitin ligase, in petals of senescing four o'clock (*Mirabilis jalapa*) flowers highly increased during the onset of visible senescence like that in *Petunia* (Xu *et al.*, 2007). Silencing of this gene also resulted in extension of flower life.

Proteasome-independent protease activity increases prior to visible senescence (Stephenson & Rubinstein, 1998; Pak & Van Doorn, 2005). The proteases are often divided into exo- and endoproteases, indicating position of the target protein from where the cleavage takes place. Endoproteases include cysteine-, serine-, aspartic-, and metalloproteases, named after the amino acid residues or the metals that are required for cleavage reaction. In gladiolus, of the total protease activity, serine proteases account for about 67–70% while cysteine proteases account for only 23–25% (Azeez *et al.*, 2007). E-64 and antipain, these are specific inhibitors used for assessing the activities of proteases, both affected cysteine proteases. Total protease activity was reduced in petals of *Hemerocallis* (Stephenson and Rubinstein, 1998) and *Gladiolus* (Arora & Singh, 2004) and in *Petunia* (Jones *et al.*, 2005) by using E-64 *in vitro*. Antipain reduced

protease activity of *Sandersonia* petals by 30% (Eason *et al.*, 2002). Feeding *Iris* petals with a membrane permeable form of E-64 also indicated that about half of the peak protease activity was due to cysteine proteases (Pak & Van Doorn, 2005). In carnation petals, a gene encoding a cysteine inhibitor, abundant at the time of flower opening, became gradually down regulated. Its mRNA had disappeared by the time of the increase in cysteine protease expression and petal wilting (Sugawara *et al.*, 2002). The cysteine protease inhibitor 2, 2-dipyridyl delayed the time to wilting in *Sandersonia* petals and prevented the senescence-associated rise in endoproteases activity (Eason *et al.*, 2002). Thus, petal senescence is accompanied by bulk non-proteasomal protein degradation, mainly in vacuoles. This process of protein degradation shows accumulation of considerable amount of amino acids during senescence. Many genes which encode protein for the proteases have also been discovered to retard this process. To some extent, this molecular approach has been proved to be involved in regulation and inhibition of proteases. Further work is also needed to prevent degradation of protein at gene level or using some cultural practices like using preservatives or inhibitors to slow down the activity of proteases.

Enzymatic Activities

An unavoidable consequence of aerobic metabolism is production of reactive oxygen species (ROS) which may be beneficial or deleterious depending upon the concentration of ROS. A high concentration

leads to damage of biomolecules and low concentration act as second messenger that mediate several responses in plants. When concentration of ROS becomes high, antioxidant system comprising of enzymatic (ascorbate peroxidase (APX), catalase (CAT), superoxide dismutase (SOD), glutathione reductase (GR) and guaiacol peroxidase (GPX) and nonenzymatic components (Ascorbate, glutathione, tocopherols, carotenoids and phenolic compounds) is there in plants to scavenge these ROS. Here, the main emphasis is given on enzymatic components like ascorbate peroxidase (APX), catalase (CAT), superoxide dismutase (SOD), glutathione reductase (GR) and guaiacol peroxidase (GPX), etc., which show antioxidant behaviour with progressive senescence. Peroxidases are one of the important enzymes found in plant material which bleaches chlorophyll in presence of H_2O_2 (Matile, 1980; Ponmeni & Mukherjee, 1997). Peroxidase activity was found to be much higher in senescent than in the young stage of leaves (Mukherjee & Rao, 1993). Enhanced peroxidase activity was associated with an increase in the level of peroxides and free radicals which react with cellular constituents (Brennan & Frenkel, 1977; Sood *et al.*, 2006). A decline in APX activity, progressive increase in SOD activity, changes in GR activity and increase in endogenous H_2O_2 were observed in *Hemerocallis* over the senescence period (Chakrabarty *et al.*, 2009). H_2O_2 is reduced by ascorbate peroxidase (APX) with the consequent oxidation of ascorbate to

dehydroascorbate while catalase converts H_2O_2 into H_2O and O_2 . The increase in SOD activity over the senescing period could be due to the over-expression of genes induced by H_2O_2 accumulation (Hossain *et al.*, 2006). In *Gladiolus* petals, the decrease in APX activity was assumed to be the prerequisite for flower senescence resulting in an increase of the endogenous H_2O_2 level. In *Iris*, APX and SOD activity decreased by the time when the tepals showed wilting, while CAT activity increased and GR activity exhibited no change (Bailly *et al.*, 2001). In daylily, the decline in APX and CAT activity along with LOX action resulted in high H_2O_2 endogenous levels during senescence (Barber & Thompson, 1980; Fukuchi-Mizutani *et al.*, 2000; Sood *et al.*, 2006).

Lipoxygenase also mediated the oxidation of polyunsaturated fatty acids and production of free radicals (Hildebrand & Hymowitz, 1982). However, the well-defined enzymatic antioxidant defense system (superoxide dismutase, SOD; ascorbate peroxidase, APX; glutathione reductase, GR) protect them against these deleterious effects by scavenging ROS. As senescence advanced, the membrane lipid peroxidation caused membrane leakage (Barber & Thompson, 1980; Fukuchi-Mizutani *et al.*, 2000; Sood *et al.*, 2006; Chakrabarty *et al.*, 2009) by the action of LOX and release free radicals.

The elevation in protease activity is among the important changes associated with the sepal senescence of *Consolidajacis* flowers (Shahri & Tahir, 2011b). Proteolytic enzymes have been divided

into several different groups depending on specific site at which they cleave target protein. Most common are cysteine proteases. Cpase are closest functional homologue to caspases in senescent plant tissue and commonly functional during petal senescence (Wagstaff *et al.*, 2002). Cpase was upregulated during petal senescence in daylily flowers (Guerrero *et al.*, 1998). Proteases are mainly classified into two categories- exo and endoproteases depending upon the cleavage site in protein. Endoproteases include cysteine-, serine- and aspartic- named after amino acid cleavage site in protein. As many as nine cysteine proteases have been isolated from senescing *Petunia* petals. In transgenic *Petunia* petals, expression of 4 cysteine proteases genes was delayed resulting in delay of petal wilting (Jones *et al.*, 2005).

The upregulation of various nucleases such as RNases and DNases also increases during senescence in many cut flowers (Winkenbach, 1970; Lesham *et al.*, 1986; Panavas *et al.*, 2000; Canetti *et al.*, 2002; Lers *et al.*, 2006). The bifunctional nucleases which are able to degrade both RNA and DNA have also been identified. Their activities and the levels of mRNA encode those increases during plant senescence (Canetti *et al.*, 2002; Perez-Amador *et al.*, 2000). Degraded nuclear DNA indicated laddering of DNA when fragments of DNA were placed on a gel found in a number of flowers like *Alstroemeria* and *gladiolus* (Wagstaff *et al.*, 2003; Yamada *et al.*, 2003). The mRNA abundance of DNase genes was also observed during senescence in petals

of *Hemerocallis* (Panavas *et al.*, 1999), *Narcissus* (Hunter *et al.*, 2004) and *Petunia* (Langston *et al.*, 2005). This part showed the formation of reactive oxygen species during stress conditions and upregulation or downregulation of various enzymes to regulate the senescence processes. Reports have confirmed that data are much less conclusive on the actual role of oxidative stress and the protective enzymatic systems with their corresponding isoenzymes in relation to progression of flower senescence in plants. However, more detailed work is needed to address this theme.

ETHYLENE

Ethylene is the major promoter of flower senescence in ethylene sensitive flowers, coordinating senescence pathways and floral abscission (Woltering & Van Doorn, 1988; Trobacher, 2009). A visible sign of senescence in ethylene sensitive flower is accompanied by sudden, transient increase in respiration resulting in burst of endogenously produced ethylene, coordinating the senescence pathways and upregulation of genes for enzymes required for senescence (Kende, 1993; Jones *et al.*, 2005; Narumi *et al.*, 2006; Ichimura *et al.*, 2009). The genes for enzymes include S-adenosylmethionine synthase, β -glucosidase, β -galactosidase, aspartic proteases, nucleases, asparagine synthetase etc. (Woodson *et al.*, 1992; Eason *et al.*, 2000; Wagstaff *et al.*, 2002; Narumi *et al.*, 2006). Ethylene biosynthesis pathway starts with conversion of S-adenosylmethionine (SAM) to ACC by upregulation of SAM synthase,

ACC synthase and ACC oxidase during ethylene sensitive petal senescence (Buffer *et al.*, 1980; Jones, 2004; Hoeberichts *et al.*, 2007). Antisense techniques prolonged life of cut carnation flowers by inhibiting the conversion of ACC to ethylene by decreasing level of ACC oxidase mRNA (Savin *et al.*, 1995; Kosugi *et al.*, 2002). In flowers of several species such as *Petunia*, carnation and orchids, senescence is mediated by pollination with evolution of ethylene following contact between pollen and stigmatic surface. Ethylene produced from pollinated stigma has been shown to be translocated via style and ovary to carnation petals triggering ethylene synthetic genes contributing to senescence (ten Have & Woltering, 1997; Shibuya *et al.*, 2000; Nukui *et al.*, 2004; Satoh *et al.*, 2005). Strong evidence for an important role for the gynoecium in carnation petal senescence was observed after gynoecium removal by hand by delaying senescence as petal ethylene unable to reach to normal ethylene level in absence of gynoecium. In unpollinated ethylene sensitive petal senescence, cells become more sensitive to their basal ethylene production, which might be related to decrease in cytokinin activity in petals. Exposure to ethylene at $0.5 \mu\text{L L}^{-1}$ or higher concentrations for 24 h markedly accelerated flower senescence, indicating that *Gentiana scabra* flowers are highly sensitive to ethylene (Shimizu-Yumoto & Ichimura, 2012). Ethylene is also known to be involved in the abscission of flower parts in plants such as *Delphinium* (Van Doorn, 2002; Ichimura *et al.*, 2009). Overexpression

of gene encoding isopentenyltransferase (*ipt*) results in high level of cytokinins extending life of petals in transgenic *Petunia* flowers. When these *ipt*-overexpressing flowers were treated with ethylene, mRNA abundance of cysteine protease gene remained low for a considerable period of time (Chang *et al.*, 2003). Treatments with 1-aminocyclopropane-1-carboxylic acid (ACC), a precursor of ethylene biosynthesis, enhanced senescence of *Hibiscus rosa-sinensis* L. flowers (Trivellini *et al.*, 2011). Eisinger (1977) and Van Staden (1995) observed cytokinin treatment delayed rise in ethylene production through decrease in ethylene sensitivity in carnation petals. Application of indole-3-acetic acid (IAA) hastened rise in ethylene production and petal wilting while it can reduce ethylene in some tissues like abscission zone (Van Staden, 1995). Abscisic acid (ABA) was also observed to enhance ethylene production and hastened petal wilting (Mayak & Dilley, 1976). If gynoecia are removed, however, induction of ethylene no longer occurs and wilting has been reported. Therefore, it is concluded that ABA acts as an inducer of ethylene only through gynoecium (Shibuya *et al.*, 2000; Nukui *et al.*, 2004). Application of GA₃ delayed rise in ethylene production and postponed petal wilting. In ethylene insensitive petal senescence, signal might be endogenous, from petal cells or may not require hormones as intermediate signals. ABA treatment in *Hemerocallis* hastened time of visible senescence (Panavas *et al.*, 1998). Treatment of GA₃ as a vase solution increased petal life span in cut daffodil

flowers (Hunter *et al.*, 2004). Depression of enzyme activities and gene expression of ACC synthase and ACC oxidase were observed in cut carnation flowers under high-temperature conditions (Yangkhamman *et al.*, 2007). Exogenous ethylene influences flower opening of cut roses (*Rosa hybrida*) by regulating the genes encoding ethylene biosynthesis enzymes (Nan *et al.*, 2005) and increases water loss, anthocyanin degradation, ethylene production and decreases vase life in *Dendrobium* orchids depending on cultivars (Almasi *et al.*, 2012). *Petunia x hybrida* over-expressing the antisense BoACS1 gene (ACC synthase) or the antisense BoACO1 gene (ACC oxidase) from broccoli showed reduced ethylene biosynthesis and delay of flower senescence (Huang *et al.*, 2007). These evidences have supported involvement of both ethylene dependent and independent pathways that lead to senescence of floral parts especially of the petals. Regarding the pattern of senescence in ethylene insensitive flowers, the data so far accumulated is scanty and more elaborate work are required to understand the ethylene independent pathway and its execution.

ABSCISIC ACID (ABA)

Another important plant hormone involved in flower senescence is ABA which accelerates senescence processes in many cut flowers (Wei *et al.*, 2003; Hunter *et al.*, 2004). ABA participates in endogenous regulation of senescence processes in rose flowers (Halevy & Mayak, 1981; Panavas *et al.*, 1998; Hunter *et al.*, 2004), and this may

be due to conversion of carotenoids to ABA. ABA accelerates senescence of cut roses by promoting petal growth and respiration, thus decreasing the carbohydrate level in the petals and triggering the chain of metabolic processes leading to aging in rose flowers (Borochoy *et al.*, 1976). ABA content decreased during bud development and increased during senescence. Several fold increase in ABA concentration was observed during the later stages of senescence which was found to be associated with a drastic reduction of flower water potential and water uptake (Kumar *et al.*, 2008a; Arrom & Munne-Bosch, 2012b). The ABA content during flower development has a well-defined trend that is common in many plant species such as squash, four o'clock, daylily and daffodil (Panavas *et al.*, 1998; Hunter *et al.*, 2004). ABA may act by increasing ethylene sensitivity as observed by exogenous application in *Hibiscus* flowers (Trivellini *et al.*, 2007, 2011), suggesting that both hormones are involved in flower senescence. A direct relation between petal ABA concentration and longevity was also observed; the higher the ABA concentration at harvest, the shorter the subsequent vase life (Muller *et al.*, 1999). In perianth of daffodils, exogenously applied ABA enhanced the premature accumulation of senescence associated transcripts in petals indicating that ABA induced the transcripts independent of ethylene which was reduced by adding GA₃ in vase solution (Hunter *et al.*, 2004). In daylily, exogenously applied ABA causes a loss of differential membrane permeability and increase in lipid

peroxidation and ion leakage (Woltering & Van Doorn, 1988).

ABA-accelerated senescence appears to be mediated through induction of ethylene synthesis, since it is not seen in flowers pretreated with ethylene (Mayak & Dilley, 1976; Ronen & Mayak, 1981; Müller *et al.*, 1999). This is because daylily flowers are ethylene-insensitive (Lay-Yee *et al.*, 1992), ABA presumably induces flower senescence independently of ethylene (Panavas *et al.*, 1998). During senescence of daffodil flowers, however, Hunter *et al.* (2002) reported that although ABA accumulated in the tepals as they senesced, it did not appear to play a signaling role in natural senescence. The increase in ABA concentrations in the tepals occurred after the induction of senescence-associated genes. They concluded that the increase in ABA content was most likely a consequence of the cellular stresses that occurred during senescence and suggested that the hormone does not trigger senescence but may help drive the process to completion. It is clear that ABA is considered as most promising growth regulator which promotes senescence and also inducer of ethylene in ethylene insensitive species but proper mechanism is not known how it induces ethylene. Involvement of ABA in both ethylene-sensitive and ethylene-insensitive flower senescence regulates distinct mechanisms which have not been fully elucidated as yet.

APPROACHES TO ENHANCE POSTHARVEST LIFE OF CUT FLOWERS

Various pre-harvest, harvest and post-harvest factors also influence the postharvest quality and longevity of cut flowers (Halevy & Mayak, 1979). The important pre-harvest factors include; cultivar, light, temperature and mineral nutrients in soils. Physical damage due to pests, diseases besides and certain physiological disorders can markedly reduce the cut flower quality. Time of harvest also influences the postharvest life of cut flowers. Flowers harvested in the afternoon retain higher amount of storage and last longer than those harvested in the morning. Harvesting of flowers is done at immature stage when they are transported at long distance and for local markets, harvested at mature stage (Bhattacharjee & De, 2003). Several postharvest factors such as temperature, relative humidity, light and ethylene, CO₂ and O₂ concentration in the environment also influence longevity of cut flowers (Salunkhe *et al.*, 1990). Therefore, post-harvest quality of ornamental plants is a crucial aspect to be considered for successful commercialization. The knowledge obtained through the study of petal PCD may be applied to the objective of producing flowers with a longer vase life (Zhou *et al.*, 2005). This goal may be achieved by preserving flower freshness during their transport, by designing new holding solutions of sugars (sucrose, glucose, trehalose, etc.) hormones, bactericides, ethanol, mineral ions and metabolic inhibitors or their combinations and by genetically modifying the flowers

through the introduction of useful genes inducing longest postharvest life. Controlled atmospheres (CA) or modified atmospheres (MAP) are the recent technologies for perishable horticultural products. MAP is an inexpensive technique involving the use of polymeric films to create a low oxygen and high carbon dioxide atmosphere within the package in order to reduce physiological changes and quality loss. It works on the same principle as controlled atmosphere while controlled atmosphere (CA) require precise control on atmospheric gases and more costly in use. But these technologies of MAP and CA are not in much practice for the ornamentals.

Carbohydrate Metabolism and Effect of Exogenous Sugars

Metabolites play a crucial role in the regulation of developmental processes as well as in response to biotic and abiotic stresses in plants (Wingler & Roitsch, 2008). Sugar provides not only energy source but also molecules controlling metabolism, development and gene expression in prokaryotes and eukaryotes (Kumar *et al.*, 2008b). The reduction in sugar status is among the important changes associated with the sepal senescence of *Consolida ajacis* L. flowers. Therefore, post-harvest life is strongly dependent on the carbohydrate status and acceptable amount of metabolic sugars that affect rate of senescence (Ho & Nichols, 1977). Senescence causes loss of complex carbohydrate and transient accumulation of soluble sugars (Buchanan-Wollaston *et al.*, 2003; Sood *et al.*,

2006). Some petals contain starch and fructan. In *Chrysanthemum* petals, both polysaccharides were degraded during petal expansion (Trusty & Miller, 1991). Young petals that reported to contain high starch concentration include *Tradescantia reflexa* (Horie, 1961); *Lilium* (Bielecki *et al.*, 2000). Waithaka *et al.* (2001) reported transfer of carbohydrate from senescent lower florets to those developed acropetally during development of inflorescence. The content of sugars in the petal tissues increased during the flower opening period and then declined during senescence in *Ranunculus asiaticus* L. (Shahri & Tahir, 2011a). Tirosh and Mayak (1988) reported that α -amylase plays an important role in mechanism of petal opening and regulates the appearance of senescence syndrome. Glucose derived from starch granules are hydrolysed via β -amylase to maltose which is exported from chloroplast, as substrate for transglucosylation reaction, producing glucose and glucosylated acceptor molecule (Smith *et al.*, 2005). Starch is present in form of granules composed of branched polymers; most of these are amylopectin-an α -1, 4, α -1, 6 linked polymer (Zeeman *et al.*, 2002). The level and translocation of carbohydrates are considered as main factor affecting the development of rose flower and also as a factor affecting postharvest life of cut flowers (Khayat & Zieslin, 1989). SPS (Sucrose phosphate synthase) also plays a key role in conversion of triose phosphate to sucrose in source leaves or may be subjected to coarse control by demand of sink tissues that cause sucrose accumulation

and observed highest level of SPS gene expression in *Oncidium goldiana* flowers (Li *et al.*, 2003).

Sucrose supplementation to cut flowers maintained their ATP levels and the movement ability for a longer time than in those kept in water (Azad *et al.*, 2008). Keeping the flower in vase solution containing sucrose has been shown to extend their vase life (Ho & Nichols, 1977; Kuiper *et al.*, 1995). Inclusion of other sugars as trehalose, mannitol and inositol in vase solution also delayed senescence in tulips (Iwaya-Inoue & Nonami, 2003; Ranwala & Miller, 2009), *Alstroemeria* (Asil & Roein, 2012) and *Gladiolus* (Otsubo & Iwaya-Inoue, 2000; Yamada *et al.*, 2003; Arora & Singh, 2006). Post-harvest application of sucrose has been reported to increase longevity of some important horticultural crops such as carnation (Verlinden & Garcia, 2004). Preservative for consumers include sugars and antimicrobial compounds that inhibit vascular occlusion (Ichimura *et al.*, 2006). At least part of sugar effect could be explained by abundance of *EIL3* mRNA, which is a transcription factor that translates ethylene signals (Hoerberichts *et al.*, 2007) and by lower levels of EIL3 protein. The presence of high sugar level was observed to promote proteasomal degradation of EIN3 (Yanagisawa *et al.*, 2003). Pun and Ichimura (2003) observed delay in ethylene biosynthesis and decreased insensitivity to ethylene. Sugars can also delay senescence in ethylene sensitive petals. In *Sandersonia*, the effect of exogenous sugar on senescence was accompanied by a delay in expression of genes involved in fatty acid and protein

remobilization (Eason *et al.*, 2002). Sucrose treatment showed varied responses in different flowers of same family. Sucrose at 0.05 and 0.2 M significantly enhanced vase life of spikes of *Aquilegia vulgaris* L. and *Consolida ajacis* L. respectively, while it is found ineffective in enhancing vase life in *Ranunculus asiaticus* (Shahri *et al.*, 2010). Sucrose addition to the vase solution exerts an effect on flower opening and senescence in cut lily flowers by altering the hormonal balance of several floral tissues among other factors (Arrom & Munne-Bosch, 2012a). Sugar was also found to have stimulatory effect in cut sweet pea flowers (Ichimura & Suto, 1999), *Eustoma* (Cho *et al.*, 2001) and roses (Hayat *et al.*, 2012) in delaying senescence. Hence, sugar metabolism plays its role actively during senescence stages, transporting mainly sucrose through the phloem. It is clear that exogenous sugars delay time of wilting symptoms during senescence; however, it is often not clear to what extent applied sugars serve to improve petal water relations by increasing the level of osmotic solutes or to delay cell death. Measurements about considerable quantities of sugars present at the time of petal wilting may also have not been detailed due to the following reasons: (a) various tissues in a petal are at different stages of senescence, (b) the cytoplasm and the vacuole may have different sugar levels, and (c) sugars are again formed at some stages during senescence. Furthermore, research is needed in this aspect of breakdown and synthesis of sugar at cellular or tissue as well as at molecular level.

Biocides

Pure water used in flower containers soon becomes contaminated with bacteria and fungi which multiply on plant tissue or debris. Microorganisms in water can cause physical plugging of cut stem and release toxic metabolites. They can evolve damaging levels of ethylene and induce hypersensitive response resulting in PCD (Alvarez, 2000). The organism responsible for production of substances such as tannins can block the conducting vessels of the stem. Some chemical substances (known as biocides) have been found helpful in keeping post-harvest life of cut flowers. 8-hydroxyquinoline (8-HQ) and 8-hydroxyquinolinecitrate (8-HQC) are commonly used biocides. They lower the pH of holding solution preventing vascular blockade of many cut flowers including cut roses (Van Doorn & Perik, 1990). 8-HQ has been known to possess strong antimicrobial properties that eliminate vascular blockage and enhance water uptake so as to maintain water balance by reducing transpiration from flower tissue (Rogers, 1973; Jowker, 2005). Other commonly used disinfectants include STS (silver thiosulphate), dichlorophen, hypochlorite and quaternary ammonium compounds (Ueyama & Ichimura, 1998; Muriithi & Ouma, 2011). A solution containing 8-HQC and sucrose is routinely used to prolong vase life in cut flowers (Lukaszewska & Skutnik, 2003). The maximum vase life and flower diameter was recorded when the cut spikes were immersed in a solution containing sucrose 2% + 8 HQC – 200 ppm + AgNO₃ 50 ppm

in all the tuberose cvs. Vaibhav, Mexican Single, Shringar, Suvasini and Prajwal (Sudagar *et al.*, 2010). Knee (2000) observed that various concentrations of biocides in a solution containing 0.2 g L⁻¹ citric acid and 10 g L⁻¹ glucose were found effective on cut roses (*Rosa hybrida* L., 'Classy'), *Alstroemeria pelegrina* L. and carnation (*Dianthus caryophyllus* L.). Elgimabi and Ahmed (2009) and Tsegaw *et al.* (2011) also reported the best result in enhancing vase life in cut roses and carnation (Edrisi *et al.*, 2012) using biocides as preservatives. Pulse treatment with HQS plus sucrose for 12 h is the most effective for improving pigmentation and use as a commercial cut flower preservative solution to delay flower senescence, enhance quality, and prolong the vase life of sweet pea (Elhindi, 2012). From this part, it is concluded that biocides or disinfectants are the important preservative to prevent growth of bacteria which results in plugging of conducting vessels for proper aeration and water uptake for better postharvest performance of cut flowers. Some disinfectants based on cetrimide and chlorhexidine are phytotoxic. Therefore, freely available, cheaper and safe materials for use to plant like sodium and calcium hypochlorite can be used. Some fungicides may also be used but their effect on vase life is found to be negative. Public concern over health and environmental issues associated materials and non-chemical methods such as use of citric acid may prove to be best for better vase life of cut flowers.

Role of Plant Growth Regulators

Auxins are needed for the initiation of floral primordia. Modifications in the auxin levels may cause abortion or different flower forms (Cheng & Zhao, 2007; McSteen, 2010). Auxins have been found to delay the senescence of cut flowers like carnation and *Petunia* (Halevy & Mayak, 1981). In addition, treatment with the synthetic auxin such as 1-naphthaleneacetic acid (NAA) is useful to reduce abscission of flower buds in roses (Halevy & Kofranek, 1976) and drop of flower-bracts in bougainvillea (Chang & Chen, 2001) and enhance postharvest life and chlorophyll b in cut *Alstroemeria hybrida* (Bagheri *et al.*, 2013). In *Theobroma cacao* flowers, a single application of NAA at anthesis, anticipates petal wilting but prevents flower abscission (Aneja *et al.*, 1999; Hasenstein & Zavada, 2001). Application of IAA is also found to hasten the rise in ethylene production and petal wilting in cut carnation flowers (Van Staden, 1995). In carnation petals, a transient increase was observed in the mRNA abundance of an Aux/IAA gene (Hoerberichts *et al.*, 2007). In potted bougainvillea postproduction, auxins delay bract and flower abscission (Meir *et al.*, 2007; Gago & Monteiro, 2011). The combination of NAA and AVG (Aminoethoxyvinylglycine, an ethylene inhibitor) extended the inflorescence vase life and longevity of opened flowers more than AVG or NAA alone. The combination also increased the number of open flowers and kept the relative fresh weight of flower stems high. Therefore, application of NAA with AVG is a highly effective treatment

for improving the postharvest life of cut *Eustoma* flowers (Yumoto & Ichimura, 2010). The auxins such as IAA and NAA strongly promoted elongation and opening. An inhibitor of auxin transport (2, 3, 5-triiodobenzoic acid, TIBA) and an inhibitor of auxin effects [α -(p-chlorophenoxy)-isobutyric acid; PCIB] inhibited elongation and opening, suggesting that endogenous auxins are among the regulators of the pedicel and ovary elongation and thus of flower opening in *Iris* (Van Doorn *et al.*, 2013).

Gibberellins are mostly used and proven growth regulators in horticulture. Most commonly used one is GA₃. Kohl and Kofranek (1957) were among the first to investigate the possible use of gibberellins in floricultural crops. According to Eason (2002), treatment of gibberellic acid, a component of certain preservative solutions has been found to delay the onset of tepal fading and wilting in *Sandersonia aurantiaca* flowers and enhanced the longevity, chlorophyll content and superoxide dismutase activities in leaf and flower samples in *Alstroemeria* (Nouri *et al.*, 2012). The use of Accel (BA+GA₄₊₇) at 25 mg L⁻¹ BA has been reported to improve flower opening in *Alstroemeria* (Muthui *et al.*, 2001). Brackmann *et al.* (2005) evaluated the effects of GA₃ on three varieties of chrysanthemums and noted the promotion of senescence of both leaves and flowers. The application of GA₃ in the field did not reduce or retard the senescence process in chrysanthemum 'Faroe' (Vieira *et al.*, 2010). This author also

studied the biochemical changes in post-harvest chrysanthemum 'Faroe' submitted to different concentrations of GA₃ applied in the field and observed increase in the level of polyamines. A concentration of 10 mg L⁻¹ GA₄₊₇ can be used to prolong vase life, delay leaf senescence and enhance post-harvest quality of *Alstroemeria* cut flowers during their transport to market (Muthui *et al.*, 2006). Gerbera cut flowers held in GA₃ at concentration of 2.5, 5 or 7.5 mg L⁻¹ had significantly higher water content in flower heads and stems, hence maintaining flower turgidity, reduction in bent neck and flower senescence and increased flower quality after 14 days of holding compared with control (Emongor, 2004). Post-harvest application of GA₃ (50 ml L⁻¹) with sucrose (50 g L⁻¹) reported to improve the fresh weight, concentration of petal sugar, activities of SOD and decreased LOX activity which delayed petal senescence and enhanced vase life of gladioli (Singh *et al.*, 2008). According to Kim and Miller (2009), spray containing GA₄₊₇ plus BA might be of commercial value in enhancing postharvest quality of tulip flowers. But concentration over 50mg L⁻¹ can lead to unwanted early senescence of mature cut tulip flowers, below this concentration would be most useful for achieving maximum delay in tepal senescence. Abadi (2010) studied the effects of different concentrations of GA₃ on growth and flowering of rose (*Rosa hybrida* cv. Poison) and found that 200 mg L⁻¹ GA₃ at pre-harvest stage improved stalk length, fresh weight and yield in rose.

Eisinger (1977) proposed that cytokinins are natural anti-senescence factors and their declining levels account for triggering increased ethylene production. Feeding carnation flowers with 6-methyl purine, an inhibitor of cytokinin oxidase/dehydrogenase, resulted in increased life span of petals suggesting that ethylene promotes inactivation of cytokinins and facilitates the senescence process (Taverner *et al.*, 2000). Chang *et al.* (2003) confirmed the role of cytokinins in flower senescence using transgenic approach. The transgenic plants over expressing IPT gene under the SAG12 promoter was found to exhibit significant delay in flower senescence and corresponding increase in the cytokinin content and less sensitivity to ethylene suggesting that the regulation of flower senescence involves the interactive operation of cytokinins and ethylene. Hoeberichts *et al.* (2007) have reported the increase in mRNA abundance of two genes encoding cytokinin oxidase/dehydrogenase during carnation petal senescence which was found to accelerate cytokinin breakdown and promote corolla senescence. Cytokinin action in plant tissue is dependent upon the type of cytokinin; one type of cytokinin occurs naturally in plants and includes zeatin, dihydrozeatin and isopentenyl adenine. Similarly, BA supplied in vase solutions extended vase life in *Grevillea* 'Sylvia' inflorescences (Setyadjit *et al.*, 2004) and in *Gerbera jamesonii* Bolus ex. Hook cv. Yanara (Jabeen *et al.*, 2008). Application of 25 and 50 mg L⁻¹ BA reduced

the weight loss, chlorosis and anthocyanin degradation in *Eustoma* flowers (Asil & Karimi, 2010). Thidiazuron (TDZ), a phenylurea compound with cytokinin like activity has been found to improve *Iris* flower opening and longevity (Macnish *et al.*, 2010). BA effectively delayed leaf yellowing and also tepal senescence in tulips (Van Doorn *et al.*, 2011). However, BA produced browning of lower stem end. This was prevented by inclusion of Ca^{2+} in solution. Taken together, it is concluded that commercial regulation of plant growth and development relies heavily on the use of synthetic plant growth regulators (PGR). Concern over the impact of these chemicals on human health and the environment has already limited their use and may limit their availability in the future. Another novel approach to modulate the action of phytohormones is by manipulation at the molecular level. Antisense ACC synthase genes blocking ACC production, or with a gene encoding an enzyme that enhance ACC breakdown, has been reported recently in tomatoes transformed plants which resulted in much reduced ethylene production and delayed fruit ripening. Similar technologies will certainly be used in the future to modify production, transport, degradation and activity of PGR.

Ethanol

Alcohols from methanol to hexanol have been tested but only ethanol has been reported to have positive response in enhancing vase life in many cut flowers. Ethanol reduces or inhibits ethylene production by preventing

activity of ACC oxidase to convert ACC to ethylene. Mechanism of action depends upon the concentration of ethanol. At low concentration, ethanol is converted in to acetaldehyde that inhibits formation of ethylene. But at high concentration, it showed negative effects on cell membrane by disrupting cell permeability. Thus, it is proposed that ethanol prevents action of ethylene and penetrating into cell membrane by binding at ethylene binding sites. Ethanol has been found to be effective in increasing vase life of carnation flowers by inhibiting ethylene biosynthesis (Heins & Blakely, 1980; Wu *et al.*, 1992). Exogenous application of ethanol has been shown to delay senescence in tomatoes (Kelly & Saltveit, 1988) and oat (*Avena sativa*) leaves (Salter & Thimann, 1980). Ethanol prevented climacteric ethylene, inhibited conversion of ACC to ethylene, interfered with the action of ACC-synthase and inhibited formation of ACC (Heins, 1980; Wu *et al.*, 1992). In *Chrysanthemum* flowers, ethanol treatment delayed the senescence of flowers and improved the quality of vase life of bluebonnet racemes (Petridou *et al.*, 2001). Pun *et al.* (1999) reported that ethanol increased the vase life of carnation flowers and cultivars showed varied responses to ethanol treatment with regard to vase life increment. Ethanol (2%), along with 2.5% sucrose, delayed senescence in *Lisianthus* flowers (Farokhzad *et al.*, 2005). Continuous treatment with 8% ethanol doubled vase life of 'White Sim' carnation (*Dianthus caryophyllus*) flowers (Wu *et al.*, 1992). In addition, 8% and 10% ethanol were also

found to be effective in delaying senescence in bougainvillea flower (Hossain *et al.*, 2007). Podd and Staden (2004) stated that ethanol, when applied at low concentration as holding solution extended vase life of cut carnation flowers. They also mentioned that low concentration of either ethanol or acetaldehyde apparently decreased the formation of ethylene by inhibiting action of ACC synthase. Treatment with ethanol delayed petal senescence of flowers, possibly through reduced sensitivity to ethylene in cut *Tweedia caerulea* (Pun *et al.*, 2013) and in carnation (Adugna *et al.*, 2012). Ethanol and aluminium sulphate treatments had the most important role in the extending longevity as well as water uptake in *Rosa hybrida* cv. Black Magic (Hajizadeh *et al.*, 2012). In *Rosa hybrida*, longest vase life and minimum ethylene production were obtained by using ethanol at 6% concentration (Imani *et al.*, 2013). Ethanol (2%) has also been reported to delay senescence in *Matricaria parthenium* L. (Kaur & Mukherjee, 2012) and *Calendula officinalis* L. (Kaur & Mukherjee, 2013)

by lowering activity of α -amylase, starch degradation, lipid peroxidation and peroxidase activity.

Mineral Ions

Ions like aluminium, boron, cobalt, calcium, copper, nickel, zinc and silver in the form of various salts at appropriate concentrations have been used to improve postharvest performance of various cut flowers (Halevy & Mayak, 1981). Calcium has been used to prolong vase life of many cut bulb flowers such as tulips and to improve the quality of carnation, roses and *Petunia* (Halevy & Mayak, 1981; Torre *et al.*, 1999; Asfanani *et al.*, 2008). It delays rose petal senescence by protecting membrane proteins and phospholipids from degradation by reducing ethylene production and maintaining solute transport (Halevy *et al.*, 2000). Cytokinin and CaCl_2 decreased the senescence percentage of petals in rose cut flowers separately and this reduction was highest at higher concentration of these substances (Mortazavi *et al.*, 2007). Combined effect of Ca as a flow resistance reducer and HQS as



Fig.1: *Calendula officinalis* flowers treated with (A) methanol 2%; (B) methanol 4%; (C) methanol 6%; (D) n-butanol 2%; (E) n-butanol 4%; (F) n-butanol 6%; (G) ethanol 2%; (H) ethanol 4%; (I) ethanol 6% and (J) control (DDW) [Courtesy of Kaur & Mukherjee, 2013].

a germicidal agent contributed to improved vase life in rose (Cortes *et al.*, 2011). Calcium acting as a second messenger in the signalling pathway leading to the induction of SOD, CAT and APX, thereby, increasing the capacity of these antioxidant enzymes to scavenge more free radicals produced in the course of senescence, leading to decrease in lipid peroxidation and increasing membrane stability, being a component of cell membranes and wall, it may also strengthen both the structures and thus delays membrane deterioration and senescence in gladiolus (Sairam *et al.*, 2011).

Aluminium as $AlCl_3$ and $Al_2(SO_4)_3$ in the holding solution has been shown to enhance the quality and longevity of cut flowers such as roses, *chrysanthemum* and tuberose due to effect of Al^{3+} to reduce pH of petal cells and stabilizing the anthocyanins (Gowda & Gowda, 1990). Aluminium sulphate is reported to acidify the holding solution, keep it free of microorganisms and also help in better opening of flower buds, thereby maintaining the freshness of cut roses (Liao *et al.*, 2001; Singh *et al.*, 2004). Ichimura *et al.* (2006) developed and tested a formulation composed of sugar, germicides and aluminium sulphate that is effective in extending the vase life of cut rose flowers.

Treatment with silver has widely used as preservative measures for cut flowers (Whitehead & De Swart, 1980). Silver is a specific inhibitor of ethylene action and has been found to inhibit ethylene induced ethylene production and respiration

(Halevy & Mayak, 1981; Rodriguez *et al.*, 1999; Binder *et al.*, 2007; Strader *et al.*, 2009). Silver thiosulphate (STS) has been used as an efficient ethylene antagonist and has been shown to increase longevity in tuberose (Abbasi & Asil, 2011). The holding solution containing nano silver and sucrose resulted in the longest vase life, best water content of the leaves and flower buds and highest fresh weight gain in roses (Hesham & Kader, 2012). Silicon and nickel also increased postharvest life by decreasing malondialdehyde content and ACC oxidase activity in cut rose flowers (Kazemi *et al.*, 2012). Maintenance of elevated carbohydrate contents and reduced level of hydrolyzing enzymes exhibited by the flowers under mineral salts and sucrose treatments can be correlated with the delay in senescence and increase in postharvest vase life of *Gerbera* flowers (Wani *et al.*, 2012). It has been proved to some extent that mineral salts of most of metals are found to be beneficial for enhancing longevity of cut flowers but the mechanism how they delay senescence is not known. Molecular approach may prove to be beneficial to understand this mechanism. Some metal salts like silver salts are highly toxic therefore, keeping public concern over health and environmental issues in mind, use of these should be avoided and nontoxic, freely available, cheaper and safe to use materials should be taken into consideration.

Metabolic Inhibitors

Ethylene is one of the factors involved in causing senescence and short vase life of

many cut flowers (Ichimura *et al.*, 2002) especially in ethylene sensitive flowers. Molecules block ethylene receptors such as cyclopropene (CP), 1-methylcyclopropene (1-MCP) and 3,3-dimethylcyclopropene (3, 3-DMCP) and that block ethylene biosynthesis α -(2-aminooxyvinyl Glycine) (AVG) and aminoxyacetic acid (AOA) have been used to prolong the vase life of ethylene-sensitive flowers (Rattanawisalanona *et al.*, 2003; Sisler & Serek, 1997; Cook *et al.*, 1985; Fujino *et al.*, 1981). A volatile compound, 1-methylcyclopropene (1-MCP), is an inhibitor of ethylene action and appears to be non-toxic. It has been reported that the vase life of various cut flowers such as carnation, *Matthiola*, *Consolida*, *Chrysanthemum*, *Anthirrinum* and *Delphinium*, can be extended by exposure to 1-MCP. Treatment with 1-MCP markedly extended the vase life of cut sweet pea as did that with STS (Ichimura *et al.*, 2002). The effect of 1-MCP has been reported to prolong the postharvest life of flowers of *Geranium* and *Gentiana* (Jones *et al.*, 2001; Ferrante *et al.*, 2006; Shimizu-Yumoto & Ichimura, 2012). Treatment with 60 nL L⁻¹ 1-MCP for 3 h with 16.47 days vase life, 2.57 mL g⁻¹ fresh weight, 2.41 mL g⁻¹ water uptake and 2.667 loss of chlorophyll index was better than other treatments (Abadi *et al.*, 2009). 1-MCP and STS extended the vase life of roses (Chamoni *et al.*, 2005) and florets and spikes of cut *Freesia* 'Cordula' (Zencirkiran, 2010). 1-MCP had a strong effect of preventing the abscission of floral buds and open flowers in mini *Phalaenopsis* cultivars (Sun *et al.*, 2009). Treatment with

2-aminoethoxyvinylglycine (AVG), an inhibitor of ACC synthase, slightly delayed flower senescence in *Hibiscus syriacus* L. and *Gentiana scabra* (Seo *et al.*, 2009; Shimizu-Yumoto & Ichimura, 2012).

Cycloheximide (CHI) is an inhibitor of de novo protein synthesis in plant tissue (Ap Rees and Bryant, 1971). It is a translational inhibitor that increased vase life of daylily (*Hemerocallis*) by inhibiting petal wilting (Lay-Yee *et al.*, 1992). CHI delays loss of proteins in *Ipomoea* by inhibiting synthesis of some specific proteases responsible for protein degradation (Sultan *et al.*, 2002). Pretreatment of *Ranunculus asiaticus* flowers with 0.05 mM CHI for 1h can be used as an effective treatment to improve postharvest longevity in this flower system (Shahri & Tahir, 2010). Aminoxyacetic acid (AOA), and fluridone, an ethylene and an ABA inhibitor, respectively, extended flower longevity (Trivellini *et al.*, 2011).

Polyamines (PA's) have been reported as effective anti-senescence agents that have ability to retard chlorophyll loss, membrane deterioration and increase in RNase and protease activities which help to slow the senescence process (Evans & Malmberg, 1989). The major polyamines comprise putrescine, spermidine and spermine, which either occurs naturally or as free bases or bound to phenolics or other low molecular weight compounds (Galston & Kaur-Sawhney, 1990). Exogenous application of spermidine has been found to transiently delay senescence of *Dianthus caryophyllus* and *Petunia hybrida* flowers which has been implicated to be due to the ability of free

spermidine to bind to the main intracellular constitutive molecules such as DNA and stabilizing their structures (Gul *et al.*, 2005; Tassoni *et al.*, 2006). On the other way, methyl-jasmonates have been found to accelerate senescence in *Petunia hybrida*, *Dendrobium* and *Phalaenopsis* (Porat *et al.*, 1993, 1995), but in *Petunia inflata*, only an earlier colour change has been reported without any promotion of petal wilting after treatment with methyl-jasmonate (Xu *et al.*, 2006). Genes encoding enzymes of the jasmonate biosynthetic pathway have been shown to be specifically expressed in floral organs (ovaries, petals and sepals) and involved in reproductive processes include maturation of anthers and release of mature pollen grains (Avanci *et al.*, 2010). The known fact that pollination triggers senescence in various flower systems and these jasmonates promote pollen maturation and release which might prove to be a mechanism for role of jasmonates in flower senescence. The role of jasmonates in the senescence of ethylene-sensitive flower systems is not clear as yet. However, more elaborate work is needed to confirm it. It is well proven that almost all metabolic inhibitors delay senescence by blocking the pathway causing senescence. Among them, some of the inhibitors like 1-MCP and AOA are very expensive that these cannot be used by floricultural industries frequently. As both are non-toxic and very expensive, they have therefore limited applications in the developing countries. Lime and potassium permanganate, which are low-cost materials, can be used to remove carbon dioxide

and ethylene, respectively in packages. These absorbers can be incorporated in sachets, labels or closure liners, or can be impregnated into the MAP film.

CONCLUSION AND FUTURE RESEARCH

Floriculture has become one of the important high value agricultural industries in many countries. However, one major obstacle for floricultural industries is an early senescence of flowers. Physiological, biochemical and morphological studies provide guidance to understand the mechanism involved during abrupt changes that occur during natural flower senescence and how it can be overwhelmed. Role of reactive oxygen species and the expression of various enzymes affecting postharvest life of cut flowers must also be well understood to control senescence of cut flowers. Adoption of inexpensive and eco-friendly products as better innovative preservation proved better in long lasting quality and decelerating all senescence promoting events with reference to the flower senescence. Biotechnological tools also contributed to raise superior postharvest traits in case of many varieties of flowers. More research work in this field is needed to make flower senescence phenomenon clear with vast scope of floriculture and use of intensive techniques to maintain them for longer period. The present review has a number of important points that are missing in research from initial to end point: (1) In ultrastructural changes, the role of the tonoplast in cell death, and the cause of its rupture, is one of the challenges

for further research on petal senescence. More detailed studies are needed at cellular or tissue level regarding this aspect; (2) Mechanism of transport and sequestration of pigments like are anthocyanins, carotenoids, and betalains inside the vacuole. Genetic studies may prove to be best tool in producing mutant variety ineffective to environment conditions; (3) Main site of phospholipid degradation and to what extent this phospholipid degradation takes place inside the vacuoles and autophagosomes and to what extent inside the membrane still has become a controversy; (4) Genes involved in degradation of macromolecule and organelle has been identified in screens but comparatively little is known about the genes whose product facilitates nutrient remobilization by degrading all these structures; (5) Less conclusive data are available on the actual role of oxidative stress and role of reactive oxygen species and how the protective enzymatic systems with their corresponding isoenzymes play its role in relation to progression of flower senescence in plants as all the enzymes act collectively; (6) Regarding the pattern of senescence in ethylene insensitive flowers, the data so far accumulated is scanty and more elaborate work is required to understand the ethylene independent pathway and its execution. (7) Use of various preservatives like sugars, biocides, mineral ions, growth hormones and metabolic inhibitors has advantages but related to public concern over health and environmental issues; they are harmful to some extent. Therefore, novel technology by manipulating role of phytohormones at molecular level may solve this problem.

REFERENCES

- Abadi, D. H. (2010). Yield and quality management of *Rosa hybrida* 'Poison' with plant growth regulators. *American-Eurasian Journal of Agriculture and Environmental Sciences*, 8, 736-740.
- Abadi, D. H., Kaviani, B., Hoor, S. S., Torkashvand, A. M., & Zarei, R. (2009). Quality management of cut carnation 'Tempo' with 1-MCP. *African Journal of Biotechnology*, 8, 5351-5357.
- Abbasi, J., & Asil, M.H. (2011). Study on prolonging the vase life of tuberose cut flowers (*Polianthes tuberosa* L.). *South Western Journal of Horticulture, Biology and Environment*, 2, 157-165.
- Aduagna, B., Belew, D., & Kassa, N. (2012). Effect of pulsing solution on postharvest performance of carnation (*Dianthus caryophyllus*) cultivars. *Trends in Horticultural Research*, 2, 8-13.
- Aleksandrushkina, N. I., Kof, E. M., Seredina, A. V., Borzov, A. A., & Vanyushin, B. F. (2008). Degradation of DNA and endonuclease activity associated with senescence in the leaves of pea of normal and aphyllous genotypes. *Russian Journal of Plant Physiology*, 55, 23-32.
- Almasi, P., Mohamed, M.T.M., Ahmad, S.H., Kadir, J., & Mirshekari, A. (2012). Postharvest responses of cut *Dendrobium* orchids to exogenous ethylene. *African Journal of Biotechnology*, 11, 3895-3902.
- Alvarez, M. (2000). Salicylic acid in machinery of hypersensitive response cell death and disease resistant, *Plant Molecular Biology*, 44, 429-442.
- Aneja, M., Gianfagna, T., & Ng, E. (1999). The roles of abscisic acid and ethylene in the abscission and senescence of cocoa flowers. *Plant Growth Regulation*, 27, 149-155.
- Ap Rees, T., & Bryant, J. A. (1971). Effects of cycloheximide on protein synthesis and respiration in disks of carrot storage tissue. *Phytochemistry*, 10, 1183-1190.

- Arora, A., & Singh, V. P. (2004). Cysteine protease gene expression and proteolytic activity during floral development and senescence in ethylene-insensitive *Gladiolus grandiflora*. *Journal of Plant Biochemistry and Biotechnology*, *13*, 123–126.
- Arora, A., & Singh, V. P. (2006). Polyols regulate flower senescence by delaying programmed cell death in *Gladiolus*. *Journal of Plant Biochemistry and Biotechnology*, *15*, 139–142.
- Arrom, L., & Munne-Bosch, S. (2012a). Sucrose accelerates flower opening and delay senescence through a hormonal effect in cut lily flowers. *Plant Science*, *41*, 88–189.
- Arrom, L., & Munne-Bosch, S. (2012b). Hormonal regulation of leaf senescence in *Lilium*. *Journal of Plant Physiology*, *169*, 1542–1550.
- Asfanani, M., Davarynejad, G. H., & Tehranifar, A. (2008). Effects of pre-harvest calcium fertilization on vase life of rose cut flowers cv. Alexander. Proc. EURASIA Sym. on Quality Management in Postharvest Systems 217. In S. Kanlayanarat *et al.* (Eds.), *Acta Horticulture*, 804, ISHS 2008.
- Asil, M. H., & Karimi, M. (2010). Efficiency of benzyladenine reduced ethylene production and extended vase life of cut *Eustoma* flowers. *Plant Omic Journal*, *3*, 199–203.
- Asil, M. H., & Roein, Z. (2012). Beneficial effect of carbohydrate solutions on postharvest characteristics of cut *Alstroemeria*. *South-Western Journal of Horticulture, Biology and Environment*, *3*, 85–98.
- Avanci, N. C., Luche, D. D., Goldman, G. H., & Goldman, M. H. S. (2010). Jasmonates are phytohormones with multiple functions, including plant defense and reproduction. *Genetics and Molecular Research*, *9*, 484–505.
- Avila-Rostant, O., Lennon, A. M., & Umaharan, P. (2010). Spathe color variation in *Anthurium andraeanum* Hort. and its relationship to vacuolar pH. *HortScience*, *45*, 1768–1772.
- Azad, A. K., Ishikawa, T., Ishikawa, T., Sawa, Y., & Shibata, H. (2008). Intracellular energy depletion triggers programmed cell death during petal senescence in tulip. *Journal of Experimental Botany*, *59*, 2085–2095.
- Azeez, A., Sane, A. P., Bhatnagar, D., & Nath, P. (2007). Enhanced expression of serine proteases during floral senescence in *Gladiolus*. *Phytochemistry*, *68*, 1352–1357.
- Bagheri, H., Hashemabadi, D., Sedaghatoor, S., Zarchini, M., & Eslami, A. (2013). Effect of naphthalene acetic acid (NAA) on vase life, chlorophyll b content and water relation of cut *Alstroemeria hybrida*. *Annals of Biological Research*, *4*, 59–61.
- Bailly, C., Corbineau, F., & Van Doorn, W. G. (2001). Free radical scavenging and senescence in *Iris* tepals. *Plant Physiology and Biochemistry*, *39*, 649–656.
- Barber, R. F. & Thompson, J. E. (1980). Senescence-dependent increase in the permeability of liposome prepared from bean cotyledon membranes. *Journal of Experimental Botany*, *31*, 1305–1313.
- Bartoli, C. G., Guaiamet, J. J., & Montaldi E. R. (1997). Ethylene production and responses to exogenous ethylene in senescing petals of *Chrysanthemum morifolium* RAM cv. Unsei. *Plant Science*, *124*, 15–21.
- Bartoli, C. G., Simontacchi, M., Montaldi E. R., & Puntarolo S. (1995). Oxidants and antioxidants during aging of chrysanthemum petals. *Plant Science*, *129*, 157–165.
- Battelli, R., Lombardi, L., Rogers, H.J., Picciarelli, P., Lorenzi, R., & Ceccarelli, N. (2011). Changes in ultrastructure, protease and caspase-like activities during flower senescence in *Lilium longiflorum*. *Plant Science*, *180*, 716–725.

- Bhattacharjee, S. K., & De, L. C. (2003). *Advanced Commercial Floriculture* (Vol. I). Aavishkar (Pub), Jaipur.
- Bialeski, R., Elgar, J., & Heyes, J. (2000). Mechanical aspects of rapid flower opening in Asiatic lily. *Annals of Botany*, *86*, 1175-1183.
- Bialeski, R. L. (1995). Onset of phloem export from senescent petals of daylily. *Plant Physiology*, *109*, 557-565.
- Bialeski, R. L., & Reid, M. S. (1992). Physiological changes accompanying senescence in ephemeral daylily flower. *Plant Physiology*, *98*, 1042-1049.
- Binder, B. M., Rodriguez, F. I., Blecker, A. B., & Patterson, S.E. (2007). The effects of group 11 transition metals, including gold, on ethylene binding to the ETR1 receptor and growth of *Arabidopsis thaliana*. *FEBS Letters*, *581*, 5105-5109.
- Biran, I., & Halevy, A. H. (1974). Effect of varying light intensities and temperature treatments applied to whole plants, or locally to leaves or flower buds, on growth and pigmentation of 'Baccara' roses. *Physiologia Plantarum*, *31*, 175-179.
- Borochoy, A., Spiegelstein, H., & Philosoph H.S. (1997). Ethylene and flower petal senescence: interrelationship with membrane lipid catabolism. *Physiologia Plantarum*, *100*, 606-612.
- Borochoy, A., Tirosh, T., & Halevy, A. H. (1976). Abscisic acid content of senescing petals on cut rose flowers as affected by sucrose and water stress. *Plant Physiology*, *58*, 175-178.
- Brackmann, A., Belle, R. A., de Freitas, S. R., & de Mello, A. M. (2005). Vase life of chrysanthemum (*Dendranthema grandiflora*) in gibberellic acid solutions. *Revista Ciesncia Rural, Santa Maria*, *35*, 1451-1455.
- Breeze, E., Wagstaff, C., Harrison, E., Bramka, I., Rogers, H., Stead, A., Thomas, B., & Buchanan-Wollaston, V. (2004). Gene expression patterns to define stages of postharvest senescence in *Alstroemeria* petals. *Journal of Plant Biotechnology*, *2*, 155-168.
- Brennan, T., & Frenkel, C. (1977). Involvement of hydrogen peroxide in regulation of senescence in pear. *Plant Physiology*, *59*, 411-416.
- Brown, J. H., Chambers, J. A., & Thompson, J. E. (1991). Acyl chain and head group regulation of phospholipid catabolism in senescing carnation flowers. *Plant Physiology*, *95*, 909-916.
- Brown, J. H., Lynch, D. V., & Thompson, J. E. (1987). Molecular species specificity of phospholipid breakdown in microsomal membranes of senescing carnation flowers. *Plant Physiology*, *85*, 679-683.
- Brown, J. H., Palyath, G., & Thompson, J. E. (1990). Influence of acyl length composition on the degradation of phosphatidylcholine by phospholipase D in carnation microsomal membranes. *Journal of Experimental Botany*, *41*, 979-986.
- Buchanan-Wollaston, V., Earl, S., Harrison, E., Mathas, E., Navabpour, S., Page, T., & Pink, D. (2003). The molecular analysis of plant senescence: a genomics approach. *Plant Biotechnology Journal*, *1*, 3-22.
- Bufler, G., Mor, Y., Reid, M. S., & Fang, S. F. (1980). Changes in 1-aminocyclopropane-1-carboxylic acid content of cut carnation flowers in relation to their senescence. *Planta*, *150*, 439-442.
- Butler, R. D., & Simon, E. W. (1971). Ultrastructural aspects of senescence in plants. In Bernad, Strehler, L. (Ed.), *Adv. Gerontol. Res* (73-129). New York: Academic Press Inc.
- Canetti, L., Lomaniec, E., Elkind, Y., & Lers, A. (2002). Nuclease activities associated with dark-induced and natural leaf senescence in parsley. *Plant Science*, *163*, 873-880.
- Chakrabarty, D., Verma, A. K., & Datta, S. K. (2009). Oxidative stress and antioxidant activity as the

- basis of senescence in *Hemerocallis* (day lily) flowers. *Journal of Horticulture and Forestry*, *1*, 113-119.
- Chamoni, E., Khalighi, A. M., Joyce, D. C., Irving, D. E., Zamani, Z. A., Mostofi, Y., & Mohsen, K. (2005). Ethylene and anti-ethylene treatment effects on cut First-Red rose. *Journal of Applied Horticulture*, *7*, 3-7.
- Chang, H., Jones, M. L., Banowetz, G. M., & Clark, D. G. (2003). Overproduction of cytokinins in petunia flowers transformed with P_{SAG12}-IPT delays corolla senescence and decreases sensitivity to ethylene. *Plant Physiology*, *132*, 2174-2183.
- Chang, Y. S., & Chen, H. C. (2001). Variability between silver thiosulfate and 1-naphthaleneacetic acid applications in prolonging bract longevity of potted bougainvillea. *Scientia Horticulturae*, *87*, 217-224.
- Chen, Z. H., Walker, R. P., Acheson, R. M., Tecsí, L. I., Wingler, A., Lea, P. J., & Leegood, R. C. (2000). Are isocitrate lyase and phosphoenolpyruvate carboxykinase involved in gluconeogenesis during senescence of barley leaves and cucumber cotyledons? *Plant and Cell Physiology*, *41*, 960-967.
- Cheng, Y., & Zhao, Y. (2007). A role for auxin in flower development. *Journal of Integrative Plant Biology*, *49*, 99-104.
- Cho, M. C., Celikel, F. G., Dodge, L., & Reid, M. S. (2001). Sucrose enhances the postharvest quality of cut flowers of *Eustoma grandiflorum* (Raf.) Shinn. *Acta Horticulturae*, *543*, 305-315.
- Collette, V. E., Jameson, P. E., Schwinn, K. E., Umaharan, P., & Davies, K. M. (2004). Temporal and spatial expression of flavonoid biosynthetic genes in flowers of *Anthurium andraeanum*. *Physiologia Plantarum*, *122*, 297-304.
- Cook, D., Rasche, M., & Eisinger, W. (1985). Regulation of ethylene biosynthesis and action in cut carnation flower senescence by cytokinins. *Journal of American Society of Horticultural Sciences*, *110*, 24-27.
- Cornah, J. E., Germain, V., Ward, J. L., Beale, M. H., & Smith, S. M. (2004). Lipid utilization, gluconeogenesis, and seedling growth in *Arabidopsis* mutants lacking the glyoxylate cycle enzyme malate synthase. *Journal of Biological Chemistry*, *279*, 42916-42923.
- Cortes, M. H., Frias, A. A., Moreno, S. G., Pina, M. M., Guzman, G. H. D. L. C., & Sandoval, S. G. (2011). The effects of calcium on postharvest water status and vase life of *Rosa hybrida* cv. Grand Gala. *International Journal of Agriculture and Biology*, *13*, 233-238.
- Dat, J. F., Vandenabeele, S., Vranova, E., Van Montagu, M., Inze, D., & Van Breusegem, F. (2000). Dual action of the active oxygen species during plant stress responses. *Cellular and Molecular Life Science*, *57*, 779-795.
- De Vetten, N. C., & Huber, D. J. (1990). Cell wall changes during the expansion and senescence of carnation (*Dianthus caryophyllus*) petals. *Physiologia Plantarum*, *78*, 447-454.
- Dela, G., Or, E., Ovardia, R., Nissim-Levi, A., Weiss, D., & Oren-Shamir, M. (2003). Changes in anthocyanin concentration and composition in 'Jaguar' rose flowers due to transient high-temperature conditions. *Plant Science*, *164*, 333-340.
- Dertinger, U., Schaza, U., & Schulze, E. D. (2003). Age-dependence of the antioxidative system in tobacco with enhanced glutathione reductase activity or senescence-induced production of cytokinins. *Physiologia Plantarum*, *119*, 19-29.
- Desai, R., Patel, R., & Mankad, A. (2012). Petal senescence in cut *Tagetes erecta* L. flowers: Role of phenolics. *International Journal of Science, Environment and Technology*, *1*, 485-490.
- Eason, J. R. (2002). *Sandersonia aurantiaca*: An evaluation of postharvest pulsing solutions to maximise cut flower quality. *New Zealand*

- Journal of Crop and Horticultural Science*, 30, 273-279.
- Eason, J. R., Johnston, J. W., de Vre, L., Sinclair, B. K., & King, G. A. (2000). Amino acid metabolism in senescing *Sandersonia aurantiaca* flowers: cloning and characterization of asparagine synthetase and glutamine synthetase cDNAs. *Australian Journal of Plant Physiology*, 27, 389-396.
- Eason, J. R., Ryan, D. J., Pinkney, T. T., & O'Donoghue, E. M. (2002). Programmed cell death during flower senescence: Isolation and characterization of cysteine proteinases from *Sandersonia aurantiaca*. *Functional Plant Biology*, 29, 1055-1064.
- Edrisi, B., Sadrpoor, A., & Saffari, V. R. (2012). Effects of chemicals on vase life of cut carnation (*Dianthus caryophyllus* L. 'Delphi') and microorganisms population in solution. *Journal of Ornamental and Horticultural Plants*, 2, 1-11.
- Eisinger, W. (1977). Role of cytokinins in carnation flower senescence. *Plant Physiology*, 59, 707-709.
- Elgimabi, M. N. & Ahmed, O. K. (2009). Effects of bactericides and sucrose-pulsing on vase life of rose cut flowers (*Rosa hybrid*). *Botany Research International*, 2, 164-168.
- Elhindi, K. M. (2012). Effects of postharvest pretreatments and preservative solutions on vase life longevity and flower quality of sweet pea (*Lathyrus odoratus* L.). *Photosynthetica*, 50, 371-379.
- Elibox, W., & Umaharan, P. (2008). Inheritance of major spathe colours in *Anthurium andraeanum* Hort. is determined by three major genes. *Hort Science*, 43, 787-791.
- Emongor, V. E. (2004). Effect of gibberellic acid on postharvest quality and vase life of gerbera cut flowers (*Gerbera jamesonii*). *Journal of Agronomy*, 3, 191-195.
- Evans, P. T., & Malmberg, R. L. (1989). Do polyamines have a role in plant development? *BioScience*, 33, 382-388.
- Farokhzad, A., Khalighi, A., Mostofi, Y., & Naderi, R. (2005). Role of ethanol in vase life and ethanol production in cut lisianthus (*Eustoma grandiflorum* Mariachii cv. Blue). *Journal of Agriculture and Social Sciences*, 1, 309-312.
- Ferrante, A., Mensuali-Sodi, A., Serra G., & Tognoni, F. (2006). Evaluation of postproduction performance of *Salvia splendens* potted plants for interiors use. *Acta Horticulturae*, 723, 415-419.
- Fobel, M., Lynch, D.V., & Thompson, J. E. (1987). Membrane deterioration in senescing carnation flowers. *Plant Physiology*, 85, 204-211.
- Fujino, D. W., Reid, M. S., & Yang, S. F. (1981). Effects of aminooxyacetic acid on postharvest characteristics of carnation. *Acta Horticulturae*, 113, 59-64.
- Fukuchi-Mizutani, M., Ishiguro, K., Nakayama, T., Utsunomiya, Y., Tanaka, Y., Kusumi, T., & Ueda, T. (2000). Molecular and functional characterization of a rose lipoxygenase cDNA related to flower senescence. *Plant Science*, 160, 129-137.
- Gago, C. M. L., & Monteiro, J. A. (2011). NAA and STS effects on bract survival time, carbohydrate content, respiration rate and carbohydrate balance of potted *Bougainvillea spectabilis* Willd. *Postharvest Biology and Technology*, 60, 235-243.
- Galston, A. W., & Davies, P. J. (1970). *Control mechanisms in plant development*. Englewood Cliffs New Jersey: Prentice Hall.
- Galston, A. W., & Kaur-Sawhney, R. (1990). Polyamines in plant physiology. *Plant Physiology*, 94, 406-410.
- Gonzalez, A. (2009). Pigment loss in response to the environment: A new role for the WD/ bHLH/

- MYB anthocyanin regulatory complex. *New Phytologist*, 182, 1-3.
- Gowda, J. V. N., & Gowda, V. N. (1990). Effects of calcium, aluminium and sucrose on vase life of gladiolus. *Crop Research*, 3, 105-106.
- Griesbach, R. J. (1992). Correlation of pH and light intensity on flower color in potted *Eustoma grandiflorum* 'Grise'. *Hort Science*, 27, 817-818.
- Griesbach, R. J. (2005). A scientific approach to breeding blue orchids: Exploring new frontiers in search of elusive flower colors. *Orchids* (W. Palm Beach, Fla.), 74, 376-379.
- Guerrero, C., de la Calle, M., Reid, M. S., & Valpusesta, V. (1998). Analysis of the expression of two thiol protease from daylily (*Heimerocallis*) during flower senescence. *Plant Molecular Biology*, 36, 565-571.
- Gul, F., Tahir, I., & Sultan, S. M. (2005). Effect of some polyamines and protein synthesis inhibitors on flower senescence in *Petunia hybrida* Vilm. cv. Tint Cascade. *Oriental Science*, 10, 117-122.
- Hajizadeh, H. S. Farokhzad, A., & Chelan, V. G. (2012). Using of preservative solutions to improve postharvest life of *Rosa Hybrid* cv. Black Magic. *Journal of Agricultural Technology*, 8, 1801-1810.
- Halevy, A., & Mayak, S. (1979). Senescence and postharvest physiology of cut flowers- part 1. *Horticultural Reviews*, 1, 204-236.
- Halevy, A., & Mayak, S. (1981). Senescence and postharvest physiology of cut flower- part 2. *Horticultural Reviews*, 3, 59-143.
- Halevy, A. H., Borochoy, A., & Torre, S. (2000). Calcium regulation of senescence in rose petals. *Physiologia Plantarum*, 107, 214-219.
- Halevy, A. H., & Kofranek, A. M. (1976). The prevention of flower bud and leaf abscission in pot roses during simulated transport. *Journal of American Society Horticultural Sciences*, 101, 658-660.
- Harborne, J. B. (1988). The flavonoids: Recent advances. In T. W. Goodwin (Ed.), *Plant Pigments* (299-343). London, UK: Academic Press Ltd.
- Hasenstein, K. H., & Zavada, M. S. (2001). Auxin modification of the incompatibility response in *Theobroma cacao*. *Physiologia Plantarum*, 112, 113-118.
- Hayat, S., Ul-Amin, N., Khan, M. A., Soliman, T. M. A., Nan, M., Hayat, K., Ahmad, I., Kabir, M. R., & Zhao, L. J. (2012). Impact of Silver thiosulfate and sucrose solution on the vase life of rose cut flower cv. Cardinal. *Advances in Environmental Biology*, 6, 1643-1649.
- Heins, R. D. (1980). Inhibition of ethylene synthesis and senescence in carnation by ethanol. *Journal of American Society Horticultural Sciences*, 105, 141-144.
- Heins, R. D., & Blakely, N. (1980). Influence of ethanol on ethylene biosynthesis and flower senescence of cut carnation. *Scientia Horticulturae*, 13, 361-369.
- Hernandez-Jimenez, M. J., Lucas, M. M., & de Felipe M. R. (2002). Antioxidant defence and damage in senescing lupin nodules. *Plant Physiology and Biochemistry*, 40, 645-657.
- Hesham, H., & Kader, A. (2012). Effects of nanosilver holding and pulse treatments, in comparison with traditional silver nitrate pulse on water relations and vase life and quality of the cut flowers of *Rosa hybrida* L. cv. 'Tineke'. *World Applied Sciences Journal*, 20, 130-137.
- Hildebrand, D. F., & Hymowitz, T. (1982). Carotene and chlorophyll bleaching by soyabean with or without seed lipoxygenase-1. *Journal of Agriculture and Food Chemistry*, 76, 249-253.
- Ho, L., & Nichols, R. (1977). Translocation of ¹⁴C-sucrose in relation to changes in carbohydrate content in rose corollas cut at different stages of development. *Annals of Botany*, 41, 227-242.

- Hoeberichts, F. A., Van Doorn, W. G., Vorst, O., Hall, R. D., & Van Wordragen, M. F. (2007). Sucrose prevents upregulation of senescence associated genes in carnation petals. *Journal of Experimental Botany*, *58*, 2873-2885.
- Hopkins, M., Taylor, C., Liu, Z., Ma, F., McNamara, L., Wang, T. W., & Thompson, J. E. (2007). Regulation and execution of molecular disassembly and catabolism during senescence. *New Phytologist*, *175*, 201-214.
- Horie, K. (1961). The behavior of the petals in the fading of the flowers of *Tradescantia reflexa*. *Protoplasma*, *53*, 377-386.
- Hortensteiner S. (2006). Chlorophyll degradation during senescence. *Annual Review of Plant Biology*, *57*, 55-77.
- Hossain, A. B. M. S., Zuliana, R., Chandran, S., Mohamed, H. A. M., & Boyce, A. N. (2007). *Bougainvillea* flower longevity and delay abscission as affected by different concentrations of ethanol. *Maejo International Journal of Science and Technology*, *2*, 234-238.
- Hossain, Z., Mandal, A. K. A. Datta S. K., & Biswas, A. K. (2006). Decline in ascorbate peroxidase activity- a prerequisite factor for tepal senescence in gladiolus. *Journal of Plant Physiology*, *163*, 186-194.
- Huang, L. C., Lai, U. L., Yang, S. F., Chu, M. J., Kuo, C. I., Tsai, M. F., & Sun, C. W. (2007). Delayed flower senescence of *Petunia hybrida* plants transformed with antisense broccoli ACC synthase and ACC oxidase genes. *Postharvest Biology and Technology*, *46*, 47-53.
- Hunter, D. A., Steele, B. C., & Reid, M. S. (2002). Identification of genes associated with perianth senescence in daffodil (*Narcissus pseudonarcissus* L. 'Dutch Master'). *Plant Science*, *163*, 13-21.
- Hunter, D. A., Ferrante, A., Vernieri, P., & Reid, M. S. (2004). Role of abscisic acid in perianth senescence of daffodil (*Narcissus pseudonarcissus* 'Dutch Master'). *Physiologia Plantarum*, *121*, 313-321.
- Hunter, D. A., Yi, M. F., Xu, X. J., & Reid, M. S. (2004). Role of ethylene in perianth senescence of daffodil (*Narcissus pseudonarcissus* L. 'Dutch Master'). *Postharvest Biology and Technology*, *32*, 269-280.
- Ichimura, K. (2010). Post-harvest physiology of cut flowers: progress and future aspects. *Bulletin of National Institute of Floricultural Science*, *10*, 11-53 (In Japanese).
- Ichimura, K., Shimizu-Yumoto, H., & Goto, R. (2009). Ethylene production by gynoecium and receptacle is associated with sepal abscission in cut *Delphinium* flowers. *Postharvest Biology and Technology*, *52*, 267-272.
- Ichimura, K., Taguchi, M., & Norikoshi, R. (2006). Extension of vase life in cut roses by treatment with glucose, isothiazolinic germicide, citric acid and aluminium sulphate solution. *Japan Agricultural Research Quarterly*, *40*, 263-269.
- Ichimura, K., Shimizu, H., Hiraya, T., & Hisamatsu, T. (2002). Effect of 1-methylcyclopropene (1-MCP) on the vase life of cut carnation, *Delphinium* and sweet pea flowers. *Bulletin of National Institute of Floricultural Science*, *2*, 1-8.
- Ichimura, K., & Suto, K. (1999). Effect of time of sucrose treatment on vase life, soluble carbohydrate concentrations and ethylene production in cut sweet pea flowers. *Plant Growth Regulation*, *28*, 117-122.
- Imani, M. H., Hashemabadi, D., Kaviani, B., & Zarchini, M. (2013). Improving water relations and postharvest quality of cut rose (*Rosa hybrida* L. cv. 'Avalanche') by ethanol. *Annals of Biological Research*, *4*, 256-259.
- Irani, N. G., & Grotewold, E. (2005). Light-induced morphological alteration in anthocyanin accumulating vacuoles of maize cells. *BMC Plant Biology*, *5*, 7.

- Iwaya-Inoue, M., & Nonami, H. (2003). Effects of trehalose on flower senescence from view point of physical status of water. *Environment Control in Biology*, *41*, 315.
- Jabeen, A., Chandrasekhar, R., & Padma, M. (2008). Effect of plant growth regulators on physical changes and microbial count during vase life of cut gerbera flowers (*Gerbera jamesonii* Bolus ex. Hook.) cv. Yanara. *Journal of Ornamental Horticulture*, *11*, 107-111.
- Jones, M. L. (2004). Changes in gene expression during senescence. In L.D. Nooden (Ed.), *Plant cell death processes* (pp. 51–71). Amsterdam: Elsevier.
- Jones, M. L., Chaffin, G. S., Eason, J. R., & Clark, D.G. (2005). Ethylene sensitivity regulates proteolytic activity and cysteine protease gene expression in petunia corollas. *Journal of Experimental Botany*, *56*, 2733-2744.
- Jones, M. L., Kim, E., & Newman, S. E. (2001). Role of ethylene and 1-MCP in flower development and petal abscission in zonal geraniums. *Hort Science*, *36*, 1305–1309.
- Jowker, M. M. (2005). Effect of different compounds on microbial population of cut ‘Shiraz narcissus’ vase solution. V. International Postharvest symposium, ISHS, *Acta Horticulture*, *682*, 1705-1708.
- Katsumoto, Y., Fukuchi-Mizutani, M., Kukui, Y., Brugliera, F., Holton, T. A., Karan, M., Nakamura, N., Yonekura-Sakakibara, K., Togami, J., Pigeaire, A., Tao, G.-Q., Nehra, N. S., Lu, C. Y., Dyson, B. K., Tsuda, S., Ashikari, T., Kusumi, T., Mason, J. G., & Tanaka, Y. (2007). Engineering of the rose flavonoid biosynthetic pathway successfully generated blue-hued flowers accumulating delphinidin. *Plant Cell Physiology*, *48*, 1589–1600.
- Kaur, P., & Mukherjee, D. (2012). Delaying postharvest senescence of *Matricaria parthenium* L. flowers using ethanol, methanol and sucrose. *Journal of tropical plant physiology*, *4*, 1-16.
- Kaur, P., & Mukherjee, D. (2013). Senescence regulation by alcohols in cut flowers of *Calendula officinalis* L. *Acta Physiologiae Plantarum*, *35*, 1853-1861.
- Kawabata, S., Ohta, S. M., Kusuvara, Y., & Sakiyama, R. (1995). Influence of low light intensities on the pigmentation of *Eustoma grandiflorum* flowers. *Acta Horticulturae*, *405*, 173-178.
- Kazemi, M., Gholami, M., & Aghdasi, S. (2012). Efficiency of silicon, nickel and acetylsalicylic acid reduced senescence and extended vase life of cut rose flowers. *Trends in Applied Sciences Research*, *7*, 590-595.
- Kelly, M. O. & Saltveit, M. E. (1988). Effect of endogenously synthesized and exogenously applied ethanol on tomato fruit ripening. *Plant Physiology*, *88*, 143-147.
- Kende, H. (1993). Ethylene biosynthesis. *Annual Review of Plant Physiology*, *44*, 283-307.
- Khayat, E., & Zieslin, N. (1989). Translocation of ¹⁴C-carbohydrate content and activity of enzymes of sucrose metabolism in rose petals at different night temperatures. *Physiologia Plantarum*, *76*, 581-585.
- Kim, H. J., & Miller, W. B. (2009). GA₄₊₇ plus BA enhance postproduction quality in pot tulips. *Postharvest Biology and Technology*, *51*, 272–277.
- Kishimoto, S., Maoka, T., Nakayama, M., & Ohmiya, A. (2004). Carotenoid composition in petals of chrysanthemum (*Dendranthemarandiflorum* (Ramat). *Phytochemistry*, *65*, 2781–278.
- Knee, M. (2000). Selection of biocides for use in floral preservatives. *Postharvest Biology and Technology*, *18*, 227–234.
- Knowles, L. Trimble, M. R., & Knowles, N. R. (2001). Phosphorus status affects postharvest

- respiration, membrane permeability and lipid chemistry of European seedless cucumber fruit (*Cucumis sativus* L.). *Postharvest Biology and Technology*, 2, 179-188.
- Kohl, H. C. Jr., & Kofranek, A. M. (1957). Gibberellin on flower crops. *California Agriculture*, 1, 9.
- Kosugi, Y., Waki, K., Iwazaki, Y., Tsuruno, N., Mochizuki, A., Yoshioka, T., Hashiba, T., & Satoh, S. (2002). Senescence and gene expression of transgenic non-ethylene-producing carnation flowers. *Journal of the Japanese Society for Horticultural Science*, 71, 638–642.
- Kuiper, D., Ribot, S. A., Van Reenen, H. S., & Marissen, N. (1995). The effect of sucrose on the flower bud opening of Madelon cut roses. *Scientia Horticulturae*, 60, 325-336.
- Kuiper, D., Van Reenen, H. S., & Ribot, S. A. (1991). Effect of gibberellic acid on sugar transport into petals of ‘Madelon’ rose flowers during bud opening. *Acta Horticulturae*, 298, 93-95.
- Kumar, N., Srivastava, G. C., & Dixit, K. (2008a). Hormonal regulation of flower senescence in roses (*Rosa hybrida* L.). *Plant Growth Regulation*, 55, 65–71.
- Kumar, N., Srivastava, G. C., & Dixit, K. (2008b). Flower bud opening and senescence in roses (*Rosa hybrida* L.) *Plant Growth Regulation*, 55, 81-99.
- Langston, B. J., Bai, S., & Jones, M. L. (2005). Increases in DNA fragmentation and induction of a senescence-specific nuclease are delayed during corolla senescence in ethylene-insensitive (etr1-1) transgenic petunias. *Journal of Experimental Botany*, 56, 15–23.
- Lay-Yee, M., Stead, A. D., & Reid, M. S. (1992). Flower senescence in daylily (*Heemerocallis*). *Physiologia Plantarum*, 86, 308-314.
- Lei, W., Geng-Guo, T., & Tong, L. (2009). Physiological, biochemical and ultrastructures of mesophyll cell changes in a senescing corolla of *Lycoris longituba*. *Journal of Zhejiang Forestry College*, 26, 498-502.
- Lers, A., Sonogo, L., Green, P. J., & Burd, S. (2006). Suppression of LX ribonuclease in tomato results in a delay of leaf senescence and abscission. *Plant Physiology*, 142, 710- 721.
- Lesham, Y. Y., Halevy, A. H., & Frenkel, C. (1986). In *Free radical and control of plant senescence* (pp. 100-116). New York: Elsevier.
- Leverentz, M. K., Wagstaff, C., Rogers, H.J., Stead, A.D., Chanasut, U., Silkowski, H., Thomas, B., Weichert, H., Feussner, I., & Griffiths, G. (2000). Characterization of novel lipoxygenase-independent senescence mechanism in *Alstroemeria* floral tissue. *Plant Physiology*, 130, 273–283.
- Leverentz, M. K., Wagstaff, C., Rogers, H.J., Stead, A.D., Chanasut, U., Sikowski, H., Thomas, B., Weichert, H., Feussner, I., & Griffiths, G. (2002). Characterization of a novel lipoxygenase independent senescence mechanism in *Alstroemeria* floral tissue. *Plant Physiology*, 130, 273-283.
- Li, C. R., Zhang, X .B., & Hew, C. S. (2003). Cloning of a sucrose-phosphate synthase gene highly expressed in flowers from the tropical epiphytic orchid *Oncidium goldiana*. *Journal of Experimental Botany*, 54, 2189–2191.
- Liao, L., Lin, Y., Huang, K., & Chen, W. (2001). Vase life of *Eustoma grandiflorum* as affected by aluminum sulfate. *Botanical Bulletin of Academia Sinica*, 42, 35-38.
- Louda, J. W., Liu, L., & Baker, E. W. (2002). Senescence and death-related alteration of chlorophylls and carotenoids in marine phytoplankton. *Organic Geochemistry*, 33, 1635-1653.
- Lukaszewska, A. J., & Skutnik, E. (2003). *Przewodnik florysty*. Wyd. SGGW, War szawa, pp. 127-138.

- Macnish, A. J., Jiang, C., & Reid, M. S. (2010). Treatment with thidiazuron improves opening and vase life of iris flowers. *Postharvest Biology and Technology*, *56*, 77–84.
- Matile, P. (1980). Catabolism of chlorophyll: involvement of peroxidase. *Zeitschrift Fur Pflanzenphysiologie*, *107*, 269-279.
- Matile, P., & Winkenbach, F. (1971). Function of lysosomes and lysosomal enzymes in senescing corolla of morning glory (*Ipomoea purpurea*). *Journal of Experimental Botany*, *23*, 759-771.
- Mayak, S., & Dilley, D. (1976). Effect of sucrose on responses of cut carnation flowers to kinetin, ethylene and abscisic acid. *Journal of American Society and Horticultural Sciences*, *101*, 583–585.
- Mazza, G. (2007). Anthocyanins and heart health. *Annali dell'Istituto Superiore di Sanita*, *43*, 369-374.
- McSteen, P. (2010). Auxin and Monocot Development. *Cold Spring Harbor Perspectives in Biology*, *2*, a001479.
- Meir, S., Kochanek, B., Glick, A., Salim, S., Lers, A., Burd, S., Philosoph-Hadas, S., & Weiss D. (2009). Reduced petal pigmentation in lisianthus (*Eustoma grandiflorum*) flowers under low light conditions is associated with decreased expression of anthocyanin biosynthesis genes. *ISHS Acta Horticulturae*, *877*: VI International Postharvest Symposium.
- Meir, S., Salim, S., Chernov, Z., & Philosoph-Hadas, S. (2007). Quality improvement of cut flowers and potted plants with postharvest treatments based on various cytokinins and auxins. *Acta Horticulturae*, *755*, 143-156.
- Meng, X., & Wang, X. (2004). Regulation of flower development and anthocyanin accumulation in *Gerbera hybrida*. *The Journal of Horticultural Science and Biotechnology*, *79*, 131-137.
- Moalem-Beno, D., Tamari, G., Leitner-Dagan, Y., Borochoy, A., & Weiss, D. (1997). Sugar-dependent gibberellin-induced chalcone synthase gene expression in *Petunia corollas*. *Plant Physiology*, *113*, 419-424.
- Moehs, C. P., Tian, L., Osteryoung, K. W., & Penna, D. (2001). Analysis of carotenoid biosynthetic gene expression during marigold petal development. *Plant Molecular Biology*, *45*, 281–293.
- Mortazavi, N., Naderi, R., Khalighi, A., Babalar, M., & Allizadeh, H. (2007). The effect of cytokinin and calcium on cut flower quality in rose (*Rosa hybrida* L.) cv. Illona. *Journal of Food, Agriculture and Environment*, *5*, 311-313.
- Mukherjee, D., & Rao, K. U. M. (1993). Alternation pattern of Hill activity, peroxidase activity and sugars of pigeonpea during maturation and senescence. *Plant Physiology and Biochemistry*, *20*, 45-48.
- Müller, F., Adori, C., & Sass, M. (2004). Autophagic and apoptotic features during programmed cell death in fat body of the tobacco hornworm (*Manduca sexta*). *European Journal of Cell Biology*, *83*, 67-78.
- Müller, R., Stummann, B. M., Andersen, A. S., & Serek, M. (1999). Involvement of ABA in postharvest life of miniature potted roses. *Plant Growth Regulation*, *29*, 143–150.
- Muriithi, K., & Ouma, G. (2011). The effect of sugar and hypochlorite on the vase life of cut roses and carnations. *Journal of Animal and Plant Sciences*, *11*, 1394-1397.
- Muthui, T. M., Emonger, V. E., & Hutchinson, M. J. (2001). Effect of Accel on vase life and postharvest quality of cut *Alstroemeria* (*Alstroemeria aurantiaca*) cut flowers. *African Journal of Science and Technology*, *2*, 82-88.
- Muthui, T. M., Emongor, V. E., & Hutchinson, M. J. (2006). The effects of gibberellin₄₊₇ on the

- vase life and flower quality of *Alstroemeria* cut flowers. *Plant Growth Regulation*, 48, 207–214.
- Nan, M., Lei, C., Wangjin, L., Hui, T., & Junping, G. (2005). Exogenous ethylene influences flower opening of cut roses (*Rosa hybrida*) by regulating the genes encoding ethylene biosynthesis enzymes. *Science in China Ser. C Life Sciences*, 48, 434-444.
- Narumi, T., Sudo, R., & Satoh, S. (2006). Cloning and characterization of a cDNA encoding a putative nuclease related to petal senescence in carnation (*Dianthus caryophyllus* L.) flowers. *Journal of the Japanese Society for Horticultural Science*, 75, 323-327.
- Nielsen, K. M., Lewis, D. H., & Morgan, E. R. (2003). Characterization of carotenoid pigments and their biosynthesis in two yellow flowered lines of *Sandersonia aurantiaca* (Hook.). *Euphytica*, 130, 25–34.
- Nouri, N., Abdossi, N., & Boojar, M. (2012). The effect of gibberellic acid and benzyladenine on post harvest quality and vase life of *Alstroemeria* cultivar Mayfair cut flowers with enzymatic assay. *International Journal of Agriculture: Research and Review*, 2, 1025-1031.
- Nowak, J., & Rudnicki, R. M. (1990). *Postharvest Handling and Storage of Cut Flowers, Florist Greens and Potted Plants (1st Edn.)*. Portland: Timber Press. ISBN-13: 978-0881921564, 210p.
- Nukui, H., Kudo, S., Yamashita, A., & Satoh, S. (2004). Repressed ethylene production in the gynoecium of long-lasting flowers of the carnation ‘White Candle’: role of the gynoecium in carnation flower senescence. *Journal of Experimental Botany*, 55, 641–650.
- O’Donoghue, E., Eason, J.R., Somerfield, S. D. & Ryan, D. A. (2005). Galactosidases in opening, senescing and water-stressed *Sandersonia aurantiaca* flowers. *Functional Plant Biology*, 32, 911–922.
- Otsubo, M., & Iwaya-Inoue, M. (2000). Trehalose delays senescence in cut gladiolus spikes. *Horticultural Sciences*, 35, 1107-1110.
- Pak, C., & Van Doorn, W. G. (2005). Delay of Iris flower senescence by protease inhibitors. *New Phytologist*, 165, 473–480.
- Panavas, T., LeVangie, R., Mistler, J., Reid, P. D., & Rubinstein, B. (2000). Activities of nucleases in senescing daylily petals. *Plant Physiology and Biochemistry*, 38, 837–843.
- Panavas, T., Pikula, A., Reid, P. D., Rubinstein, B., & Walker, E. L. (1999). Identification of senescence-associated genes from daylily petals. *Plant Molecular Biology*, 40, 237–248.
- Panavas, T., Reid, P. D., & Rubinstein, B. (1998). Programmed cell death of daylily petals: activities of wall-based enzymes and effects of heat shock. *Plant Physiology and Biochemistry*, 36, 379–388.
- Perez-Amador, M. A., Abler, M. L., De Rocher, E. J., Thompson, D. M., Van Hoof, A., Le Brasseur, N. D., Lers, A., & Green, P. J. (2000). Identification of BFN1, a bifunctional nuclease induced during leaf and stem senescence in Arabidopsis. *Plant Physiology*, 122, 169-179.
- Petridou, M., Voyiatzi, C., & Voyiatzi, D. (2001). Methanol, ethanol and other compounds retards leaf senescence and improve the vase life and quality of cut chrysanthemum flowers. *Postharvest Biology and Technology*, 23, 79-83.
- Pistelli, L., Perata, P., & Alpi, A. (1992). Effects of leaf senescence on glyoxylate cycle enzyme activities. *Australian Journal of Plant Physiology*, 19, 723-729.
- Plaut, Z., Zieslin, N., Grawa, A., & Gazit, M. (1979). The response of rose plants to evaporative cooling: Flower production and quality. *Scientia Horticulturae*, 11, 183-190.
- Podd, L. A., & Staden, J. V. (2004). The role of ethanol and acetaldehyde in flower senescence and fruit

- ripening. *Journal of Plant Growth Regulation*, 26, 183-189.
- Ponmeni, G., & Mukherjee, D. (1997). Kinetin induced alteration in senescence of red gram leaves. *Indian Journal of Plant Physiology*, 2, 250-251.
- Porat, R., Borochoy, A., & Halevy, A. (1993). Enhancement of *Petunia* and *Dendrobium* flower senescence by jasmonic acid methyl ester is via the promotion of ethylene production. *Plant Growth Regulation*, 13, 297-301.
- Porat, R., Reiss, N., Atzorn, R., Halevy, A. H., & Borochoy, A. (1995). Examination of the possible involvement of lipoxygenase and jasmonates in pollination-induced senescence of *Phalaenopsis* and *Dendrobium* flowers. *Physiologia Plantarum*, 94, 205-210.
- Prochazkova, D., Sairam, R. K., Srivastava, G. C., & Singh, D. V. (2001). Oxidative stress and antioxidant activity as the basis of senescence in maize leaves. *Plant Science*, 161, 765-771.
- Pun, U. K., & Ichimura, K. (2003). Role of sugars in senescence and biosynthesis of ethylene in cut flowers. *Japan Agricultural Research Quarterly*, 37, 219-224.
- Pun, U. K., Niki, T., & Ichimura, K. (2013). Ethanol reduces sensitivity to ethylene and delays petal senescence in cut *Tweedia caerulea* flowers. *Plant Growth Regulation*, 69, 125-130.
- Pun, U. K., Rowe, R. N., Rowarth, J. S., Barnes, M. F., Dawson, C. O., & Heyes, J. A. (1999). Influence of ethanol on climacteric senescence in five cultivars of carnation. *New Zealand Journal of Crop and Horticultural Sciences*, 27, 69-77.
- Ranwala, A. P., & Miller, W. B. (2009). Comparison of the dynamics of non-structural carbohydrate pools in cut tulip stems supplied with sucrose or trehalose. *Postharvest Biology and Technology*, 52, 91-96.
- Rattanawisalanona, C., Ketsa S., & Van Doorn W. G. (2003). Effect of aminooxyacetic acid and sugars on the vase life of *Dendrobium* flowers. *Postharvest Biology and Technology*, 29, 93-100.
- Rodriguez, F. I., Esch, J.J., Hall, A. E., Binder, B. M., Schaller, G. E., & Bleecker, A. B. (1999). A copper cofactor for the ethylene receptor ETR1 from *Arabidopsis*. *Science*, 283, 996-998.
- Rogers, H. J. (2006). Programmed cell death in floral organs: How and why do flowers die? *Annals of Botany*, 97, 309-315.
- Rogers, H. J. (2012). Is there an important role for reactive oxygen species and redox regulation during floral senescence? *Plant, Cell and Environment*, 35, 217-233.
- Rogers, M. N. (1973). An historical and critical review of postharvest physiology research on cut flowers. *Hort. Science*, 8, 189-194.
- Ronen, M., & Mayak, S. (1981). Interrelationship between abscisic acid and ethylene in the control of senescence processes in carnation flowers. *Journal of Experimental Botany*, 32, 759-765.
- Roy, P., Roy, S. P., Mitra, A., & Kulkarni, A. P. (1994). Superoxide generation by lipoxygenase in presence of NADH and NADPH. *Biochimica et Biophysica Acta*, 1214, 171-179.
- Rubinstein, B. (2000). Regulation of cell death in flower petals. *Plant Molecular Biology*, 44, 303-318.
- Sairam, R. K., Vasanthan, B., & Arora, A. (2011). Calcium regulates *Gladiolus* flower senescence by influencing antioxidative enzymes activity. *Acta Physiologiae Plantarum*, 33, 1897-1904.
- Salter, S. O., & Thimann, K. V. (1980). The influence of aliphatic alcohols on leaf senescence. *Plant Physiology*, 66, 395-399.
- Salunkhe, F. B., Bhat, N. R., & Desai, B. B. (1990). *Postharvest Biotechnology of Flowers and Ornamental Plants*. Berlin, Heidelberg: Springer-Verlag.

- Sankhla, N., Mackay, W. A., & Davis, T. D. (2005). Corolla abscission and petal color in cut *Phlox* flower heads: Effects of sucrose and thidiazuron. Proc. VIIIth IS Postharvest Phys. Ornamentals. In N. Marissen *et al.* (Eds.), *Acta Horticulturae*, 669, ISHS.
- Satoh, S., Nukui, H., & Inokuma, T. (2005). A method for determining the vase life of cut spray carnation flowers. *Journal of Applied Horticulture*, 7, 8-10.
- Savin, K. W., Baudinette, S. C., Graham, M. W., Michael, M. Z., Nugent, G. D., Lu, C. Y., Chandler, S. F., & Cornish, E. C. (1995). Antisense ACC oxidase RNA delays carnation petal senescence. *HortScience*, 30, 970-972.
- Seo, S-G., Kang, S-W., Shim, I-S., Kim, W., & Fujihara, S. (2009). Effects of various chemical agents and early ethylene production on floral senescence of *Hibiscus syriacus* L. *Plant Growth Regulation*, 57, 251-258.
- Setyadjit, Joyce, D. C., Irving, D. E., & Simons, D. H. (2004). Effects of 6-benzyladenine treatments on the longevity of harvested *Grevillea* 'Sylvia' inflorescences. *Plant Growth Regulation*, 43, 9-14.
- Shahri, W. (2011). Senescence: Concept and Synonyms. *Asian Journal of Plant Sciences*, 10, 24-28.
- Shahri, W., & Tahir, I. (2010). Effect of cycloheximide on senescence and post-harvest performance of *Ranunculus asiaticus* L. flowers. *Pakistan Journal of Botany*, 42, 3577-3585.
- Shahri, W., & Tahir, I. (2011a). Flower development and senescence in *Ranunculus asiaticus* L. *Journal of Fruit and Ornamental Plant Research*, 19, 123-131.
- Shahri, W., & Tahir, I. (2011b). Physiological and biochemical changes associated with flower development and senescence in *Consolida ajacis* Nieuwl cv. Violet Blue. *Frontiers of Agriculture in China*, 5, 201-208.
- Shahri, W., Tahir, I., Islam, S. T., & Ahmad, M. (2010). Response of some ornamental flowers of family Ranunculaceae to sucrose feeding. *African Journal of Plant Sciences*, 4, 346-352.
- Shahri, W., Tahir, I., Islam, S.T., & Bhat, M.A. (2011). Physiological and biochemical changes associated with flower development and senescence in so far unexplored *Helleborus orientalis* Lam. cv. Olympicus. *Physiology and Molecular Biology of Plants*, 17, 33-39.
- Shibuya, K., Yoshioka, T., Hashiba, T., & Satoh, S. (2000). Role of the gynoecium in natural senescence of carnation (*Dianthus caryophyllus* L.) flowers. *Journal of Experimental Botany*, 51, 2067-2073.
- Shimizu-Yumoto, H., & Ichimura, K. (2012). Effects of ethylene, pollination, and ethylene inhibitor treatments on flower senescence of gentians. *Postharvest Biology and Technology*, 63, 111-115.
- Shoji, K., Miki, N., Nakajima, N., Momonoi, K., Kato, C., & Yoshida, K. (2007). Perianth bottom-specific blue colour development in tulip cv. Murasakizuisho requires ferric ions. *Plant Cell Physiology*, 48, 243-251.
- Shvarts, M., Borochoy, A., & Weiss, D. (1997). Low temperature enhances petunia flower pigmentation and induces chalcone synthase gene expression. *Physiologia Plantarum*, 99, 67-72.
- Singh, A., Kumar, J., & Kumar, P. (2008). Effects of plant growth regulators and sucrose on postharvest physiology, membrane stability and vase life of cut spikes of gladiolus. *Plant Growth Regulation*, 55, 221-229.
- Singh, K., Singh, P. J., & Kumar, R. (2004). Effect of some chemical treatments on keeping quality of cut roses. *Advances in Horticultural Sciences*, 18, 161-167.
- Sisler, E. C., & Serek, M. (1997). Inhibitors of ethylene responses in plants at receptor level:

- Recent developments. *Physiologia Plantarum*, *100*, 577-582.
- Smith, A. M., Zeeman, S. C., & Smith, S. M. (2005). Starch degradation in *Arabidopsis*. *Annual Review of Plant Biology*, *56*, 73-98.
- Smith, M., & Butler, R. D. (1971). Ultrastructural aspects in *Cucumis sativus* with particular reference to chromoplasts. *Protoplasma*, *73*, 1-13.
- Smith, M. T., Saks, Y., & Van Staden, J. (1992). Ultrastructural changes in petals of senescing flowers of *Dianthus caryophyllus* L. *Annals of Botany*, *69*, 277-285.
- Sood, S., Vyas, D., & Nagar, P.K. (2006). Physiological and biochemical studies during flower development in two rose species. *Scientia Horticulturae*, *108*, 390-396.
- Spundova, M., Popelkova, H., Ilik, P., Skotnica, J., Novotny, R., & Naus, J. (2003). Ultrastructural and functional changes in the chloroplasts of detached barley leaves senescing under dark and light conditions. *Journal of Plant Physiology*, *160*, 1051-1058.
- Stead, A., & Van Doorn, W. G. (1994). Strategies of flower senescence: a review. In R. J. Scott and A. D. Stead (Eds.), *Molecular and Cellular Aspect of Plant Reproduction* (pp. 215-238). Cambridge.
- Stephenson, P., & Rubinstein, B. (1998). Characterization of proteolytic activity during senescence in day lilies. *Physiologia Plantarum*, *104*, 463-473.
- Strader, L. C., Beisner, E. R., & Bartel, B. (2009). Silver ions increase auxin efflux independently of effects on ethylene response. *The Plant Cell*, *21*, 3585-3590.
- Sudagar, I. P., Sankarnarayanan, R., & Aruna, P. (2010). Effect of chemicals in increasing the vase life of tuberose cultivars. *The Asian Journal of Horticulture*, *4*, 421-423.
- Sugawara, H., Shibuya, K., Yoshioka, T., Hashiba, T., & Satoh, S. (2002). Is a cysteine proteinase inhibitor involved in the regulation of petal wilting in senescing carnation (*Dianthus caryophyllus* L.) flowers? *Journal of Experimental Botany*, *53*, 407-413.
- Sultan, S. M., Tahir, I., Arif, M., & Farooq, S. (2002). Cycloheximide spray treatment soon after opening prolongs longevity of detached morning glory (*Ipomoea tricolor*) flowers. *Journal of Plant Biology*, *29*, 105-118.
- Sun, Y., Christensen, B., Liu, F., Wang, H., & Muller, R. (2009). Effects of ethylene and 1-MCP (1-methylcyclopropene) on bud and flower drop in mini *Phalaenopsis* cultivars. *Plant Growth Regulation*, *59*, 83-91.
- Tassoni, A., Accettulli, P., & Bagni, N. (2006). Exogenous spermidine delays senescence of *Dianthus caryophyllus* flowers. *Plant Biosystematics*, *140*, 107-114.
- Taverner, E., Lethem, D. S., Wang, J., & Cornish, E. (2000). Inhibition of carnation petal inrolling by growth retardants and cytokinins. *Australian Journal of Plant Physiology*, *27*, 357-362.
- Ten Have, A., & Woltering, E. J. (1997). Ethylene biosynthetic genes are differentially expressed during carnation (*Dianthus caryophyllus* L.) flower senescence. *Plant Molecular Biology*, *34*, 89-97.
- Thompson, J. (1988). The molecular basis for membrane deterioration during senescence. In L.D. Noodén and A.C. Leopold (Eds.), *Senescence and Aging in Plants* (pp. 51-83). New York: Academic.
- Thompson, J. E., Mayak, S., Shinitzky, M., & Halevy, A. H. (1982). Acceleration of membrane senescence in cut carnation flower by treatment with ethylene. *Plant Physiology*, *69*, 859-863.
- Tirosh, T., & Mayak, S. (1988). Changes in starch content during development of carnation petals. *Journal of Plant Physiology*, *113*, 361-363.

- Torre, S., Borochoy, A., & Halevy, A. H. (1999). Calcium regulation in senescence in rose petals. *Physiologia Plantarum*, 107, 214-219.
- Tripathi, S. K., & Tuteja, N. (2007). Integrated signaling in flower senescence. *Plant signaling and Behavior*, 2, 437-44.
- Trivellini, A., Ferrante, A., Lucchesini, M., Mensuali-Sodi, A. Vernieri, P., Tognoni, F., & Serra, G. (2007). Ethylene and abscisic acid interaction during hibiscus (*Hibiscus rosa-sinensis* L.) flower development and senescence. *Advances in Plant Ethylene Research*, 2, 75-79.
- Trivellini, A., Ferrante, A., Vernieri, P., Mensuali-Sodi, A., & Serra, G. (2011). Effects of promoters and inhibitors of ethylene and ABA on flower senescence of *Hibiscus rosa-sinensis* L. *Journal of Plant Growth Regulation*, 30, 175-184.
- Trobacher, C. P. (2009). Ethylene and programmed cell death in plants. *Botany*, 87, 757-769.
- Trusty, S. E., & Miller, W. B. (1991). Postproduction carbohydrate levels in pot chrysanthemums. *Journal of the American Society for Horticultural Science*, 116, 1013-1018.
- Tsegaw, T., Tilahun, S., & Humphries, G. (2011). Influence of pulsing biocides and preservative solution treatment on the vase life of cut rose (*Rosa hybrida* L.) varieties *Ethiopian Journal of Applied Sciences and Technology*, 2, 1-18.
- Uddin, A. F. M. J., Hashimoto, F., Kaketani, M., Shimizu, K., & Sakata, Y. (2001). Analysis of light and sucrose potencies on petal coloration and pigmentation of lilyanthis cultivars (*in vitro*). *Scientia Horticulturae*, 89, 73-82.
- Ueyama, S., & Ichimura, K. (1998). Effects of 2-hydroxy-3-ionene chloride polymer on vase life of rose flowers. *Postharvest Biology and Technology*, 14, 65-70.
- Vaknin, H., Bar-Akiva, A., Ovadia, R., Ada Nissim-Levi, A., Forer, I., Weiss, D., & Oren-Shamir, M. (2005). Active anthocyanin degradation in *Brunfelsia calycina* (yesterday-today-tomorrow) flowers. *Planta*, 222, 19-26.
- Van Doorn, W. G. (2002). Effect of ethylene on flower abscission: A survey. *Annals of Botany* (London), 89, 689-693.
- Van Doorn, W. G. (2011). Classes of programmed cell death in plants, compared to those in animals. *Journal of Experimental Botany*, 62, 4749-4761.
- Van Doorn, W. G., & Perik, R. R. J. (1990). Hydroxyquinoline citrate and low pH prevent vascular blockages in stems of cut rose flowers by reducing number of bacteria. *Journal of American Society and Horticultural Sciences*, 115, 979-981.
- Van Doorn, W. G., & Woltering, E. (2004). Senescence and programmed cell death: Substance or semantics? *Journal of Experimental Botany*, 55, 2147-2153.
- Van Doorn, W. G., & Woltering, E. J. (2008). Physiology and molecular biology of petal senescence. *Journal of Experimental Botany*, 59, 453-480.
- Van Doorn, W. G., Dole, I., Celikel, F. G., & Harkema, H. (2013). Opening of *Iris* flowers is regulated by endogenous auxins. *Journal of Plant Physiology*, 170, 161-164.
- Van Doorn, W. G., Perik, R. R. J., Abadie, P., & Harkema, H. (2011). A treatment to improve the vase life of cut tulips: Effects on tepal senescence, tepal abscission, leaf yellowing and stem elongation. *Postharvest Biology and Technology*, 61, 56-63.
- Van Doorn, W. G., Balk, P. A., van Houwelingen, A. M., Hoeberichts, F. A., Hall, R. D., Vorst, O., van der Schoot, C., & van Wordragen, M. F. (2003). Gene expression during anthesis and senescence in iris flowers. *Plant Molecular Biology*, 53, 845-863.

- Van Staden, J. (1995). Hormonal control of carnation flower senescence. *Acta Horticulturae*, 405, 232-239.
- Verlinden, S., & Garcia, J. J. V. (2004). Sucrose loading decreases ethylene responsiveness in carnation (*Dianthus caryophyllus* cv. White Sim) petals. *Postharvest Biology and Technology*, 31, 305-312.
- Vieira, M. R. S., Teixeira da Silva, J. A., Lima, G. P. P., & Vianello, F. (2010). Changes in polyamine, total protein and total carbohydrate content and peroxidase activity during the lifetime of chrysanthemum 'Faroe'. *Floriculture and Ornamental Biotechnology*, 48, 52.
- Voleti, S. R., Singh, V. P., Arora, A., Singh, N., & Kushwaha, S.R. (2000). Physiology of flower senescence in floriculture crops. In A. Hemantaranjan (Ed.), *Advances in Plant Physiology* (pp. 423-439). Jodhpur, India. Jodhpur: Scientific Publishers.
- Wagstaff, C., Leverentz, M.K., Griffiths, G., Thomas, B., Chanasut, U., Stead, A. D., & Rogers, H. J. (2002). Cysteine protease gene expression and proteolytic activity during senescence of *Alstroemeria* petals. *Journal of Experimental Botany*, 53, 233-240.
- Wagstaff, C., Malcom, P., Arfan, R., Leverentz, M.K., Griffith, G., Thomas, B., Stead, A.D., & Rogers, H. (2003). Programmed cell death (PCD) processes begin extremely early in *Alstroemeria* petal senescence. *New Phytologist*, 160, 49-59.
- Waithaka, K., Dodge, L. L., & Reid, M. S. (2001). Carbohydrate traffic during opening of gladiolus florets. *Journal of Horticultural Science and Biotechnology*, 76, 20-24.
- Wani, M., Saha, S., Bidwai, J., & Khetmalas, M. (2012). Changes in carbohydrate levels and associated enzyme activities during post harvest vase life of *Gerbera jamesonii* cv. Danalin flowers as influenced by mineral salts. *Journal of Horticulture Letters*, 2, 08-11.
- Wei, Z., Zhang, H., Gu, Z. P., & Zhang, J. J. (2003). Cause of senescence of nine sorts of flowers. *Acta Botanica Sinica*, 33, 429-436.
- Weiss, D. (2000). Regulation of flower pigmentation and growth: multiple signaling pathways control anthocyanin synthesis in expanding petals. *Physiologia Plantarum*, 110, 152-157.
- Whitehead, C. S., & De Swardt, G. H. (1980). The inhibitory effect of silver ions on certain metabolic processes after uptake and distribution in different floral parts of carnations. *Agroplanta*, 2, 61-64.
- Wiemken, V., Wiemken, A., & Matile, P. (1976). Physiologie der Blüten von *Ipomoea tricolor* (Cav.): Untersuchungen an abgeschnittenen Blüten und Gewinnung eines Phloemexudates. *Biochemie und Physiologie der Pflanzen*, 169, 363-376.
- Wilhelmova, N., Domingues, P. M. D. N., Srbova, M., Fuksova, H., & Wilhelm, J. (2006). Changes in non-polar aldehydes in bean cotyledons during aging. *Biologia Plantarum*, 50, 559-564.
- Wills, R., McGlasson, B., Graham, D., & Joyce, D. (1998). *Postharvest: An Introduction to the Physiology and Handling of Fruit, Vegetables and Ornamentals* (4th Edn.). Sydney: UNSW Press, pp. 1-262.
- Wingler, A., & Roitsch T. (2008). Metabolic regulation of leaf senescence: interactions of sugar signalling with biotic and abiotic stress responses. *Plant Biology* (Stuttg), 1, 50-62.
- Winkenbach, F. (1970). Zum Stoffwechsel der aufblühenden und Welkenden Korolle der Prunkwilde *Ipomea purpurea*. *Berichte der Schweizerischen Botanischen Gesellschaft*, 80, 374-406.
- Woltering, E. J., & Van Doorn, W. G. (1988). Role of ethylene in senescence of petals- morphological and taxonomical relationships. *Journal of Experimental Botany*, 39, 1605-1616.

- Woodson, W. R., Park, K. Y., Drory, A., Larsen, P. B. & Wang, H. (1992). Expression of ethylene biosynthetic pathway transcripts in senescing carnation flowers. *Plant Physiology*, *99*, 526–532.
- Wu, M. J., Zacarias, L., Saltveit, M. E., & Reid, M. S. (1992). Alcohols and carnation senescence. *Horticultural Sciences*, *27*, 136-138.
- Xu, Y., Ishida, H., Reisen, D., & Hanson, M.R. (2006). Upregulation of a tonoplast-localized cytochrome P450 during petal senescence in *Petunia inflata*. *BMC Plant Biology*, *6*.
- Xu, X., Jiang, C. Z., Donnelly, L., & Reid, M. S. (2007). Functional analysis of a RING domain ankyrin repeat protein that is highly expressed during flower senescence. *Journal of Experimental Botany*, *58*, 3623–3630.
- Xu, X., Jiang, C., & Reid, M. S. (2008). Functional analysis of a putative ubiquitin ligase that is highly expressed during flower senescence. *Journal of Experimental Botany* (in press).
- Yamada, T., Ichimura, K., Kanekatsu, M., & Van Doorn, W. G. (2007). Gene expression in opening and senescing petals of morning glory (*Ipomoea nil*) flowers. *Plant Cell Reports*, *26*, 823–835.
- Yamada, T., Ichimura, K., & Van Doorn, W. G. (2006). DNA degradation and nuclear degeneration during programmed cell death in petals of *Antirrhinum*, *Argyranthemum* and *Petunia*. *Journal of Experimental Botany*, *57*, 3543–3552.
- Yamada, T., Takatsu, Y., Manabe, T., Kasumi, M., & Marubashi, W. (2003). Suppressive effect of trehalose on apoptotic cell death leading to petal senescence in ethylene-insensitive flowers of gladiolus. *Plant Science*, *164*, 213-221.
- Yamaguchi, T., Fukada-Tanaka, S., Inagaki, Y., Saito, N., Yonekura-Sakakibara, K., Tanaka, Y., Kusumi, T., & Iida, S. (2001). Genes encoding the vacuolar Na⁺/H⁺ exchanger and flower coloration. *Plant Cell Physiology*, *42*, 451–461.
- Yanagisawa, S., Yoo, S. D., & Sheen, J. (2003). Differential regulation of EIN3 stability by glucose and ethylene signalling in plants. *Nature*, *425*, 521–525.
- Yangkhamman, P., Koji, T., Ichimura, K., & Fukai, K. (2007). Depression of enzyme activities and gene expression of ACC synthase and ACC oxidase in cut carnation flowers under high-temperature conditions. *Plant Growth Regulation*, *53*, 155–162.
- Yao, N., Eisfelder, B. J., Marvin, J., & Greenberg, J. T. (2004). The mitochondrion - an organelle commonly involved in programmed cell death in *Arabidopsis thaliana*. *Plant Journal*, *40*, 596-610.
- Yoshida, K. (2003). Molecular regulation of leaf senescence. *Current Opinion in Plant Biology*, *6*, 79-84.
- Yoshida, K., Kawachi, M., Mori, M., Maeshima, M., Kondo, M., Nishimura, M., & Kondo, T. (2005). The involvement of tonoplast proton pumps and Na⁺(K⁺)/H⁺ exchangers in the change of petal colour during flower opening of morning glory, *Ipomoea tricolor* cv. Heavenly Blue. *Plant Cell Physiology*, *46*, 407–415.
- Yoshida, K., Kondo, T., Okazaki, Y., & Katou, K. (1995). Cause of blue petal colour. *Nature*, *373*, 291.
- Yoshida, K., Toyama-Kato, Y., Kameda, K., & Kondo, T. (2003). Sepal color variation of *Hydrangea macrophylla* and vacuolar pH measured with a proton-selective microelectrode. *Plant Cell Physiology*, *44*, 262–268.
- Yumoto, H., & Ichimura, K. (2010). Combination pulse treatment of 1-naphthaleneacetic acid and aminoethoxyvinylglycine greatly improves postharvest life in cut *Eustoma* flowers. *Postharvest Biology and Technology*, *56*, 104–107.

- Zamani, S., Kazemi, M., & Aran, M. (2011). Postharvest life of cut rose flowers as affected by salicylic acid and glutamine. *World Applied Science Journal*, 12, 1621-1624.
- Zeeman, S. C., Pilling, E., Tiessen, A., Kato, L., Donald, A. M., & Smith, A. M. (2002). Starch synthesis in *Arabidopsis*; granule synthesis, composition and structure. *Plant Physiology*, 129, 516-529.
- Zencirkiran, M. (2010). Effects of 1-MCP (1-methylcyclopropene) and STS (silver thiosulphate) on the vase life of cut *Freesia* flowers. *Scientific Research and Essays*, 5, 2409-2412.
- Zhou, Y., Wang, C.H., Ge, H., Hoeberichts, F. A., & Visser, P. B. (2005). Programmed cell death in relation to petal senescence in ornamental plants. *Acta Botanica Sinica*, 47, 641-650.

