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Review

Programmed senescence of plant organs

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Abstract

The senescence of plant organs associated with reproductive development has been studied extensively during the past century, and it has long been recognized that this type of death is internally programmed. The regulation of organ senescence as well as its biochemical and genetic determinants has been an historically rich area of research. Certain plant hormones have been implicated as regulators or modulators of organ senescence and many of the biochemical pathways associated with the senescence syndrome have been elucidated. The genetic basis of organ senescence has also been well established by the identification of mutations that impair the senescence program and recently, transgenic plants have been used to critically determine the role of specific enzymes and hormonal signals in mediating programmed senescence of plant organs. Here, we review the current understanding of the processes that regulate leaf, flower and fruit senescence, emphasizing the role that programmed organ senescence plays in the adaptive fitness of plants.

Keywords: survivorship; fruit ripening; organ senescence

Abbreviations: PCD, programmed cell death; ACC, aminocyclopropane carboxylic acid; *Nr*, *Never ripe*

Introduction

Programmed cell death (PCD) is an integral part of many aspects of plant development including xylogenesis, sex determination, leaf abscission and the hypersensitive response to pathogen infection. In each of these cases, specific groups of cells within a larger population of living cells are triggered to die. This feature of cell-specificity of the death process clearly implicates specific development signals and cell-autonomous biochemical processes targeting the execution of individual cells. There is a great deal of current research attempting to identify common features of these PCD processes, including the role of nucleosomal fragmentation, cysteine proteases and lipid-based signal molecules (Gilchrist, 1997; Dangl, 1997). It now appears likely that these cases of cell-autonomous PCD may be analogous to apoptotic cell death that has been characterized in many animal cells.

In this article we will address senescence of whole plants or of plant organs which differ fundamentally from the processes listed above in that entire populations of cells die synchronously. In most cases, organ senescence and even whole-plant senescence represents a PCD process because it is genetically determined, under the control of endogenous regulators and requires the ordered activation of specific gene expression. In many cases, the endogenous signals and genetic determinants of organ senescence are well characterized, although the understanding of the biochemical mechanisms that lead to cell death is only fragmentary. The best characterized cases of programmed organ senescence are associated with plant reproductive development, particularly leaf senescence that is triggered by flowering, flower senescence that is triggered by pollination and the ripening and senescence of fruit.

Leaf senescence

The concept that whole plant and organ senescence is a normal developmental process and under endogenous control is often attributed to Molisch (1938) who theorized that senescence was endogenously induced by depletion of material in assimilatory portions of the plant. This idea of 'exhaustive death' has been largely discredited since leaf senescence is triggered in many plants in the absence of nutrient mobilization to developing fruit (Leopold *et al*, 1959; Thomas and Stoddart, 1980). However, the association of leaf senescence with the transition from vegetative to reproductive development is widely accepted as being integral to the reproductive strategy of plants. Leopold (1961) described plant senescence in terms of survivorship curves (Pearl and Miner, 1935; Figure 1). The first of these curves describes the death of populations of individuals as a simple stochastic process in which death occurs randomly as would be observed in populations that experience death at a constant rate due to predation, stress or other external forces (curve A). Most animals in the wild belong to this group. The second curve describes a population in which the probability of death increases after a certain period of time. This pattern describes the survivorship of many domesticated animals, man, and many perennial plants which exhibit a low mortality rate until about half the maximal age and then rapidly decline (curve B). These populations appear to become more susceptible to external forces as a result of progressive deterioration of endogenous defense systems (aging), with death often resulting from disease. The third curve describes a population in which death of a population occurs catastrophically (curve C). In the absence of an external life-threatening

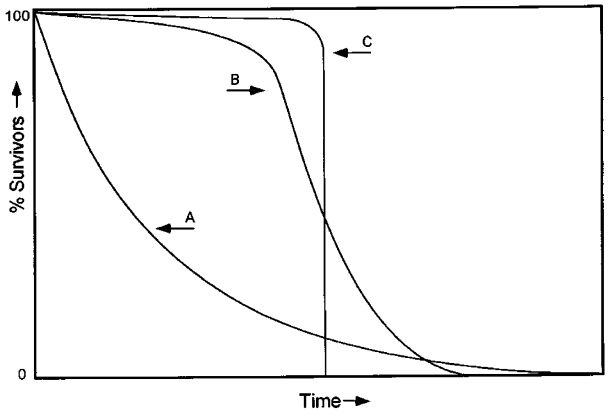

Figure 1 Generalized survivorship curves for populations in which (A) death occurs randomly as a result of external forces; (B) the probability of death increases after a certain period of time; and (C) all the members of the population die at once as a result of an endogenous programmed developmental event. [Redrawn from Pearl and Miner (1935)].

force, this pattern of death reflects the most extreme case of senescence in which all the members of the population die at once as a result of an endogenously programmed developmental event. This pattern is typical of annual plant species and does not result from an external catastrophe but from an endogenous signal and program that synchronizes the death of large populations of plants immediately following the reproductive developmental phase. There are many examples of this pattern of monocarpic senescence including most of the annual crops such as wheat and soybean.

The analysis of survivorship curves clearly illustrates that many plant species exhibit a life history strategy in which death and sudden population turnover are programmed events. Leopold (1961) reasoned that programmed senescence in populations that do not experience significant predation may serve as a mechanism to facilitate population turnover and he proposed that programmed plant senescence serves as a catalyst for evolutionary change. In addition to providing fuel for natural selection, programmed plant senescence also provides the basis for nutrient remobilization to seeds and to facilitate seed dispersal through ripening and ultimate deterioration of fruit. Thus, the life history of annual plants differs significantly from that of many animals and programmed senescence plays a central role in plant reproductive strategies and potentially in their long-term evolutionary fitness. The process of whole plant senescence is dominated by programmed death of leaves, and there has been intense research to identify the signals triggering the death response and to identify genetic and biochemical determinants of leaf senescence.

Endogenous signals regulating leaf senescence

It has long been recognized that development of various plant organs is influenced by other plant parts (Sachs, 1882) and it is generally accepted that this developmental control is mediated by plant hormones. In general terms, ethylene and abscisic acid are commonly thought of as promoters of senescence while cytokinin, auxin and gibberellins have been considered senescence inhibitors. This is a deceptively simplified view, however, and many cases exist where a senescence promoting or inhibiting hormone can have the opposite effect, depending on the tissue type, stage of development and/or interactions with other hormones. The role of the plant hormone ethylene in regulating leaf senescence has been rigorously tested by the analysis of transgenic plants suppressed for ethylene production and in mutants defective in ethylene perception. Senescence is delayed in both transgenic ethylene deficient plants (Picton *et al*, 1993a; John *et al*, 1995) and ethylene insensitive mutant plants (Bleecker *et al*, 1988; Grbic and Bleecker, 1995). However, plants that constitutively overproduce ethylene did not exhibit premature leaf senescence (Lanahan *et al*, 1994; Guzman and Ecker, 1990) as might be expected from the senescence promoting effect of exogenously applied ethylene. Grbic and Bleecker (1995) resolved this apparent discrepancy by hypothesizing that the development of leaves in the presence of high constitutive levels of ethylene reduces the endogenous sensitivity of the plant to ethylene-induced senescence. The available data convincingly shows that ethylene is not necessary for leaf senescence to occur but that it may act to modulate the timing of this process.

Other plant hormones have been shown to promote senescence in a variety of organs and cell types, such as the yellowing of detached leaves promoted by application of abscisic acid (El-Antably *et al*, 1967) or methyl jasmonate (Ueda and Kato, 1980). The responses to these hormones, however, are often mediated by ethylene, implying a secondary role, in many cases, as promoters of senescence. A recently identified class of plant hormones, the brassinosteroids, appear to be involved in a number of developmental processes and may promote leaf and chloroplast senescence (Li *et al*, 1996), although a direct role remains to be demonstrated.

The retardation of leaf senescence by cytokinin has been well-documented. Richmond and Lang (1957) reported that kinetin reduced the amount of protein lost from detached leaves, Mothes and Engelbrecht (1961) showed that treatment of a region of a senescing leaf with kinetin prevented the loss of chlorophyll and caused the accumulation of amino acids in that region, and Osborne (1962) demonstrated that the application of kinetin to senescing leaves resulted in an increase in the rate of RNA or protein synthesis relative to untreated leaves. Recently, analysis of transgenic plants with increased levels of cytokinins in specific tissues normally undergoing senescence provided overwhelming evidence implicating cytokinin as a senescence-retarding hormone. Expression of a bacterial gene encoding isopentenyl transferase under the control of a senescence-regulated promoter in transgenic tobacco resulted in elevated cytokinin levels and greatly retarded leaf senescence (Gan and Amasino, 1995). In addition, rates of photosynthesis in these transgenic plants were maintained at levels comparable to young non-senescing leaves, demonstrating that endogenous cytokinin can regulate the senescence process. In tomato fruit, the expression of the same bacterial gene under the control of a fruit-specific promoter resulted in

altered ripening properties and the authors concluded that, as in leaves, elevated cytokinin levels prevented the progression of fruit senescence (Martineau *et al*, 1994).

Taken together, it is clear that a number of hormones influence leaf senescence and that the process can be manipulated by modifying endogenous hormone levels. However, the endogenous signals that initiate leaf senescence or transmit a 'reproductive signal' from the developing ovary and fruit to the vegetative plant organs are unknown and are unlikely to be the hormones described above.

Genetic and biochemical determinants of leaf senescence

Leaf senescence is genetically determined and delayed or non-senescent mutants of soybean, fescue and sorghum have been identified (Pierce *et al*, 1984; Thomas, 1987; Duncan *et al*, 1981). Among these mutants a class of non-senescent sorghum genotypes exhibited altered cytokinin levels, suggesting that one mechanism of genetic control of senescence is altered hormone biosynthesis or transport (Ambler *et al*, 1987). Other non-senescent mutants appear more limited in their scope, with the fescue non-yellowing (NY) mutant being defective in chlorophyll loss but essentially unchanged in other parameters of senescence (Thomas and Stoddart, 1975; Thomas, 1987).

In addition to the genetic evidence that specific lesions can block the process of senescence it is also well-established that protein synthesis is required for senescence to proceed (Thomas, 1976; Shibaoka and Thimann, 1970; De Vecchi, 1971; Martin and Thimann, 1972; Makovetzki and Goldschmidt, 1976; Yu and Kao, 1981) and recent results have identified the induction of a number of specific mRNAs that are associated with leaf senescence. *In vitro* translation of mRNA from senescing leaves demonstrated that the amount of translatable mRNA encoding the small subunit of RuBPCase and other photosynthetic proteins decline during senescence, while several other mRNAs increase (Speirs and Brady, 1981; Skadsen and Cherry, 1983; Hensel *et al*, 1993). This result demonstrated that there is both positive and negative regulation of gene expression associated with leaf senescence. Strong evidence that leaf senescence involves an ordered disassembly of cell components has been revealed by the identification of mRNAs whose expression is senescence-regulated. The cDNA clones representing senescence-regulated mRNAs encode a number of proteins with activities targeted towards the breakdown of cellular components, including ribonucleases (Taylor *et al*, 1993), polyubiquitin (Garbarino *et al*, 1995), cysteine proteases (Hensel *et al*, 1993; Lohman *et al*, 1994; Smart *et al*, 1995; Buchanan-Wollaston and Ainsworth, 1997) and cell wall hydrolases (King *et al*, 1995). In addition, mRNA encoding glutamine synthetase is senescence-regulated, suggesting a role of this enzyme in the remobilization of protein nitrogen into a commonly translocated amino acid, glutamine (Kamachi *et al*, 1992). The identification and cataloguing of a large number of genes whose expression is regulated by senescence clearly indicates that the program of cell death is highly coordinated and serves a valuable role in the remobilization of scarce nutrients to surviving plant organs, such as seeds. A classification scheme for the entire set of known senescence-regulated genes has been recently proposed and provides a conceptual framework for assembling the complex processes that contribute to programmed senescence of leaves (Buchanan-Wollaston, 1997).

Flower senescence

The flower is one of the most ephemeral of plant organ systems because it is specialized for the specific functions of pollen dispersal and reception, after which time many individual floral organs senesce while others develop further to form seeds and fruit. This transition in flower function involves the programmed senescence of the petals and sepals. Perianth senescence of some flowers occurs as part of a temporal program with the petals and sepals senescing strictly as a function of age. For example, daylily flowers senesce 12–18 h after flower opening (Lukaszewski and Reid, 1989; Lay-Yee *et al*, 1992). The endogenous signals that regulate age-dependent petal senescence are completely uncharacterized, although the process is accompanied by the regulated expression of a suite of genes, some of which are functionally related to those associated with leaf senescence. Other flowers, such as petunia, gradually senesce over a period of days after flower opening, but this process is accelerated by pollination. In still other flowers perianth senescence is absolutely dependent on pollination and in these cases the external stimuli and endogenous signals that regulate programmed senescence have been examined in detail (O'Neill *et al*, 1993; O'Neill and Nadeau, 1997; O'Neill, 1997).

Endogenous signals regulating pollination-induced flower senescence

In flowers whose senescence is pollination-dependent or pollination-accelerated, including petunia, carnation, cyclamen and orchids, senescence and the pollination event is accompanied by a sudden and rapid increase in endogenous ethylene production (Nichols, 1966, 1968; Bufler *et al*, 1980; Halevy *et al*, 1984; Nichols *et al*, 1983; Whitehead *et al*, 1983; Hall and Forsyth, 1967; Porat *et al*, 1994). Indeed, it has been known for over 30 years that senescence of certain orchid flowers was accompanied by ethylene evolution and that senescence could be induced by application of exogenous ethylene (Akamine, 1963). Most importantly, the effects of exogenous ethylene could be mimicked by pollination, which indicated that ethylene played a central role in signaling the onset of programmed flower senescence.

Because the stigma is the initial site of perception of pollination, the initial pollination signal must be transduced and translocated to promote senescence processes in the distal organs of the flower, such as the perianth. A detailed characterization of transduction of the pollination signal in orchid flowers indicates that the initial pollination event promotes the synthesis of the immediate precursor of ethylene, aminocyclopropane-carboxylic acid (ACC), in the stigma by induction of expression of an ACC synthase gene

Table 1 Genes that are upregulated during fruit ripening and encode proteins of known function or exhibit significant sequence homology to proteins of known function. Proteins are grouped into five general categories based on their proposed cellular function. Multiple references indicate the cloning of a single gene sequence

Homology or function	Plant	Reference
Cell Wall Disassembly		
polygalacturonase	tomato	Slater *et al*, 1985; DellaPenna *et al*, 1986; Lincoln *et al*, 1987; Sheehy *et al*, 1987
polygalacturonase	avocado	Dopico *et al*, 1993; Kutsunai *et al*, 1993
polygalacturonase	peach	Lester *et al*, 1994
polygalacturonase	apple	Atkinson, 1994
polygalacturonase (MPG1)	melon	Hadfield, Rose and Bennett, unpublished
polygalacturonase (MPG2)	melon	Hadfield, Rose and Bennett, unpublished
polygalacturonase (MPG3)	melon	Hadfield, Rose and Bennett, unpublished
pectin methylesterase	tomato	Hall *et al*, 1994; Turner *et al*, 1996
β-galactosidase	tomato	Carey *et al*, 1995
β-galactosidase	apple	Ross *et al*, 1994
endoglucanase	avocado	Christofferson *et al*, 1984
endoglucanase (TCel 1)	tomato	Lashbrook *et al*, 1994
endoglucanase (TCel 2)	tomato	Lashbrook *et al*, 1994
endoglucanase	pepper	Ferrarese *et al*, 1995; Harpster *et al*, 1997
expansin	tomato	Rose *et al*, 1997
expansin	melon	Rose *et al*, 1997
expansin	strawberry	Rose *et al*, 1997
xyloglucan endotransglycosylase	tomato	Arrowsmith & de Silva, 1995
xyloglucan endotransglycosylase	melon	Rose and Bennett, unpublished
Ethylene Biosynthesis and Signal Transduction		
ACC[1] synthase	zucchini	Sato and Theologis, 1989
ACC synthase	tomato	Van Der Straeten *et al*, 1990
ACC synthase	apple	Dong *et al*, 1991; Lay-Yee and Knighton, 1995
ACC synthase	melon	Miki *et al*, 1995
ACC oxidase	tomato	Slater *et al*, 1985
ACC oxidase	avocado	McGarvey *et al*, 1992
ACC oxidase	apple	Dong *et al*, 1992; Ross *et al*, 1992
ACC oxidase	melon	Balague *et al*, 1993
S-adenosyl-L-methionine synthetase	kiwifruit	Whittaker *et al*, 1995
oxoglutarate-dependent dioxygenase	tomato	Lincoln *et al*, 1987
ethylene receptor	tomato	Wilkinson *et al*, 1995b
Pigmentation		
phytoene synthase (TOM5)	tomato	Slater *et al*, 1985
phytoene synthase	pepper	Romer *et al*, 1993
phytoene synthase	melon	Karvouni *et al*, 1995
phytoene desaturase	tomato	Aracri *et al*, 1994; Pecker *et al*, 1992
phytoene desaturase	pepper	Hugueney *et al*, 1992
GGPP[2] synthase	pepper	Kuntz *et al*, 1992
chalcone synthase	strawberry	Wilkinson *et al*, 1995a
Stress and Pathogen Related		
heat shock protein (TOM111)	tomato	Slater *et al*, 1985
heat shock protein (TOM66)	tomato	Fray *et al*, 1990
proteinase inhibitor (E17)	tomato	Lincoln *et al*, 1987
proteinase inhibitor (2A11)	tomato	Pear *et al*, 1989
cysteine proteinase inhibitor	avocado	Dopico *et al*, 1993
thaumatin	avocado	Dopico *et al*, 1993
thaumatin	cherry	Fils-Lycaon *et al*, 1996
endo-chitinase	avocado	Dopico *et al*, 1993
plant defensin	pepper	Meyer *et al*, 1996
major latex protein	pepper	Pozueta-Romero *et al*, 1995
major latex protein	melon	Aggelis *et al*, 1997
metallothionenin	kiwifruit	Ledger and Gardner, 1994
pollen allergen	apple	Atkinson *et al*, 1996
Metabolism		
acid invertase	tomato	Klann *et al*, 1992; Elliott *et al*, 1993
short chain alcohol dehydrogenase	tomato	Picton *et al*, 1993b
alcohol dehydrogenase	tomato	Longhurst *et al*, 1994
histidine decarboxylase	tomato	Slater *et al*, 1985
glutamate decarboxylase	tomato	Gallego *et al*, 1995
arginine decarboxylase	tomato	Rastogi *et al*, 1993
UDPGT[3]	tomato	Picton *et al*, 1993b
membrane intrinsic protein	tomato	Slater *et al*, 1985
lipoxygenase (tomloxA)	tomato	Ferrie *et al*., 1994

Table continued overleaf

Table 1 continued. . . .

Homology or function	Plant	Reference
Metabolism		
lipoxygenase (tomloxB, LOX)	tomato	Ferrie *et al*, 1994, Kausch and Handa, 1995
alternative oxidase	mango	Cruz-Hernandez and Gomez-Lim, 1995
40S ribosomal protein s12 (RNase)	strawberry	Wilkinson *et al*, 1995a
annexin	strawberry	Wilkinson *et al*, 1995a
ubiquitin conjugating enzyme	tomato	Picton *et al*, 1993b
vacuolar processing protease	citrus	Alonso and Granell, 1995
cytochrome P450	avocado	Bozak *et al*, 1990

[1]l-aminocyclopropane-1-carboxylic acid. [2]geranylgeranyl pyrophosphate. [3]UDP-glucosyl and glucuronosyl transferase

(Bui and O'Neill, submitted). ACC produced in the stigma may be oxidized directly to ethylene or potentially translocated to distal floral organs where it serves as substrate for ACC oxidase, the enzyme responsible for its conversion to ethylene. This model suggests that interorgan transduction of the pollination signal occurs by translocation of a hormone precursor, but that the endogenous signal initiating programmed senescence of flower organs is ethylene. This conclusion is supported by tomato mutants defective in ethylene perception in which the flowers do not senesce (Lanahan *et al*, 1994). In this regard, the regulation of flower senescence differs fundamentally from the regulation of leaf senescence, where ethylene appears to only modify the primary signals that underlie the senescence process.

Genetic and biochemical determinants of flower senescence

Like leaf senescence, flower organ senescence also serves to remobilize nutrients from the petals to the developing ovary and the process is active, being brought about by changes in gene expression (Lawton *et al*, 1989, 1990; Woodson, 1987; Borochov and Woodson, 1989). A number of cDNAs have been isolated from senescing carnation petals which were shown to be regulated by ethylene and correlated with the senescence process (Lawton *et al*, 1989, 1990). Two of the senescence-related cDNAs encode a β-galactosidase (SR12) and a glutathione s-transferase (SR5; Meyer *et al*, 1991). The β-galactosidase in senescing flower petals is most likely involved in cell wall disassembly that accompanies many senescence processes and it is speculated that the glutathione s-transferase acts in detoxification of lipid and DNA hydroperoxides associated with senescence-induced oxidative processes (Sylvestre *et al*, 1989). Thus, as with leaf senescence, biochemical events associated with macromolecular disassembly of floral organs are regulated at the level of gene expression in senescing petals. Because the senescence program of floral organs has been primarily investigated in those flowers whose senescence is pollination-induced and ethylene-regulated, it is not yet known what other signals may regulate the programmed senescence of other types of flowers and whether the biochemical processes of macromolecular disassembly are similar.

Fruit ripening and senescence

Fruit ripening is perhaps the most extensively characterized senescing plant organ. This process is a highly ordered developmental event that results in significant modification of the tissue composition prior to its ultimate deterioration and death. The ripening process promotes dissemination of seeds and so plays an obvious evolutionary role in expanding the geographic distribution of plant species. While fruit ripening promotes seed dissemination by animal consumption, subsequent senescence of the fruit results in increased pathogen susceptibility and seed dispersal following microbial tissue maceration.

In spite of its ecological role in seed dispersal, ripening has been primarily studied because of its economic significance in regulating the quality and longevity of fruit for human consumption. Like flower senescence, fruit ripening can be subdivided into at least two categories. The best studied category includes fruit such as tomato and bananas, where ripening is ethylene-regulated and is associated with a large increase in respiration. The large increase in respiration was originally called the climacteric and fruit in this category are thus referred to as 'climacteric fruit'. In contrast, ripening of nonclimacteric fruit is ethylene independent and is not associated with increased respiration. While it is clear that both classes of fruit undergo similar processes involving changes in pigmentation and disassembly of macromolecular cell components, very little is known about the endogenous regulators of nonclimacteric fruit ripening while a great deal is known about the regulation of climacteric fruit ripening.

Endogenous signals regulating fruit ripening

Ripening fruit have historically provided an excellent model system to study the hormonal control of senescence and early experiments showed that ethylene is the major factor contributing to the ripening and senescence of climacteric fruit. Evidence that ethylene was a positive senescence factor included the early observations that senescence of unripe fruit was stimulated by exposure to combustion fumes (Sievers and True, 1912) and that the active component of these fumes was ethylene (Denny, 1924). The possibility that ethylene in combustion fumes mimicked natural regulators of ripening and senescence became clear somewhat later, when ethylene was identified as a natural metabolite produced by some fruit (Gane, 1934) and when it was observed that ethylene depletion by hypobaric storage inhibited fruit ripening (Burg and Burg, 1965). The possible role of ethylene as the primary regulator of fruit ripening and senescence was a powerful concept that motivated a detailed elucidation of the ethylene biosynthetic pathway in plants which culminated in

1978 with the identification of the immediate precursor of ethylene, 1-aminocyclopropane-carboxylic acid (ACC; Adams and Yang, 1979). Shortly thereafter, the enzyme responsible for ACC biosynthesis, ACC synthase, was identified and shown to be a primary site of regulation of the ethylene biosynthetic pathway (Kende and Boller, 1981). The role of ethylene as the primary regulator of fruit ripening was then firmly established by suppression of ethylene production in transgenic fruit expressing antisense ACC-synthase or ACC-oxidase genes which inhibited ripening in a manner that was fully reversible by external application of ethylene (Oeller *et al*, 1991; Hamilton *et al*, 1990).

While it is clear that ethylene is a major determinant of ripening and senescence of climacteric fruit, there appear to be as yet unidentified endogenous signals that are associated with fruit maturation and enhanced sensitivity to ethylene. Because it is now known that expression of the ethylene receptor gene, *Nr*, is developmentally regulated in fruit (Payton *et al*, 1996), it is possible that competence to perceive ethylene and enter the ripening and senescence process may depend on elaboration of components of an ethylene response pathway.

Genetic and biochemical determinants of fruit ripening

The identification and characterization of tomato mutants impaired in their ability to ripen demonstrated that fruit ripening is genetically determined (Tigchelaar *et al*, 1978). Many of these mutants are altered only in their ability to synthesize carotenoids, with other ripening changes occurring normally. Other mutants, however, are affected in multiple ripening properties and recently, it has been shown that a lesion in the gene encoding the ethylene receptor is the molecular basis of the ethylene-insensitive mutant, *Never-ripe* (*Nr*) (Wilkinson *et al*, 1995b). The prevention of fruit ripening by inhibitors of protein synthesis further supports the genetic basis of ripening and suggests that it involves an orderly process of disassembly that relies on the completion of a defined set of biochemical reactions (Frenkel *et al*, 1968; Brady *et al*, 1970).

One of the most prominent biochemical processes in ripening and senescing fruit is the disassembly of macromolecules, particularly cell wall polysaccharides (Brady, 1988; Fischer and Bennett, 1991). Changes in translatable mRNA populations were shown to occur in ripening fruit (Speirs *et al*, 1984; Christoffersen *et al*, 1982) and a number of these products were identified, among the first of which were the cell wall hydrolases endo-β-1,4-glucanase (Christoffersen *et al*, 1984) and polygalacturonase (DellaPenna *et al*, 1986). In addition, a large number of other ripening-regulated genes have been identified including those that encode ethylene biosynthetic enzymes, lycopene biosynthetic enzymes and proteases. Table 1 summarizes all of the ripening-regulated genes with a putative function that have been identified to date. A large number of ripening-regulated genes of unknown function have also been catalogued and because of the extensive research in this area, the list of ripening-regulated genes grows longer on a regular basis. The precise function of each ripening-regulated gene has only been tested in a few cases, usually by the production of transgenic plants in which expression of a specific gene is suppressed by the action of antisense or sense transgenes or by its ectopic expression (Sheehy *et al*, 1988; Oeller *et al*, 1991; Smith *et al*, 1988; Giovannoni *et al*, 1989). By this process, the major role of ACC synthase in regulating the induction of fruit ripening and the role of phytoene synthase in regulating production of carotenoids has been demonstrated (Oeller *et al*, 1991; Bramley *et al*, 1992). In contrast, experiments to suppress the expression of single cell wall hydrolase genes have had only minor effects on the overall ripening process, suggesting that the complex biochemical and structural changes that accompany ripening and senescence result from the cooperative action of a potentially large number of gene products (Sheehy *et al*, 1988; Smith *et al*, 1988; Giovannoni *et al*, 1989; Lashbrook and Bennett, unpublished result).

Conclusions

Many plant organs undergo programmed senescence which is genetically determined, controlled by endogenous regulators and carried out by the ordered activation of specific gene expression. The unique life history strategies of annual plants provides an unusual example of programmed senescence of an entire organism and, indeed, entire plant populations. Although the cellular mechanisms of programmed senescence of entire plant organs are not known, there are a number of commonalities in the endogenous signals that trigger the death process and in the classes of genes that are activated to carry out the 'execution'. This suggests that the cellular mechanisms of programmed organ senescence may also share common features.

Unlike other PCD processes in plants, where cell death is restricted to a few cells involved in a hypersensitive response or to developing tracheary elements, organ senescence proceeds uniformly in large contiguous cell populations. In this regard, the propagation of signals that regulate programmed organ senescence may differ fundamentally from signals that elicit cell death in other developmental responses where death is confined to restricted cell populations. Thus, a fundamental issue for future research will be to understand how endogenous signals of cell death are propagated in contiguous cell populations of senescing organs and how this process differs from cell death processes that become restricted to a few cells within the same organ.

References

Adams DO and Yang SF (1979) Ethylene biosynthesis: identification of 1-aminocyclopropane-1-carboxylic acid as an intermediate in the conversion of methionine to ethylene. Proc. Natl. Acad. Sci. USA 76: 170–174

Akamine EK (1963) Ethylene production in fading *Vanda* orchid blossoms. Science 140: 1217–1218

Ambler JR, Morgan PW and Jordan WR (1987) Genetic regulation of senescence in a tropical grass. In Plant Senescence: Its Biochemistry and Physiology, in Thomson WW, Nothnagel EA and Huffaker RC, (eds.) Rockville: Amer. Soc. Plant Physiol pp. 43–55

Aggelis A, John I, Karvouni Z and Grierson D (1997) Characterization of two cDNA clones for mRNAs expressed during ripening of melon (*Cucumis melo* L.) fruits. Plant Mol. Biol. 33: 313 – 322

Alonso JM and Granell A (1995) A putative vacuolar processing protease is regulated by ethylene and also during fruit ripening in *Citrus* fruit. Plant Physiol. 109: 541 – 547

Aracri B, Bartley GE, Skolnik PA and Giuliano G (1994) Sequence of the phytoene desaturase locus of tomato. Plant Physiol. 106: 789

Arrowsmith DA and de Silva J (1995) Characterisation of two tomato fruit-expressed cDNAs encoding xyloglucan *endo*-transglycosylase. Plant Mol. Biol. 28: 391 – 403

Atkinson RG (1994) A cDNA clone for endopolygalacturonase from apple. Plant Physiol. 105: 1437 – 1438

Atkinson RG, Perry J, Matsui T, Ross GS and Macrae EA (1996) A stress-, pathogenesis-, and allergen-related cDNA in apple fruit is also ripening-related. New Zealand J. Crop Hortic. Sci. 24: 103 – 107

Balague C, Watson CF, Turner AJ, Rouge P, Picton S, Pech JC and Grierson D (1993) Isolation of a ripening and wound-induced cDNA from *Cucumis melo* L. encoding a protein with homology to the ethylene-forming enzyme. Eur. J. Biochem. 212: 27 – 34

Bleecker AB, Estelle MA, Somerville C and Kende H (1988) Insensitivity to ethylene conferred by a dominant mutation in *Arabidopsis thaliana*. Science 241: 1086 – 1089

Borochov A and Woodson RH (1989) Physiology and biochemistry of flower petal senescence. Hortic. Rev. 11: 15 – 43

Bozak KR, Yu H, Sirevag R and Christoffersen RE (1990) Sequence analysis of ripening-related cytochrome P-450 cDNAs from avocado fruit. Proc. Natl. Acad. Sci. USA 87: 3904 – 3908

Brady CJ (1988) Nucleic acid and protein synthesis. In Senescence and aging in plants in Nooden LD, Leopold AC (eds.) San Diego: Academic Press Inc. 147 – 179

Brady CJ, Palmer JK, O'Connell PBH and Smillie RM (1970) An increase in protein synthesis during ripening of the banana fruit. Phytochem. 9: 1037 – 1047

Bramley P, Teulieres C, Blain I, Bird C and Schuch W (1992) Biochemical characterisation of transgenic tomato plants in which carotenoid synthesis has been inhibited through the expression of antisense RNA to pTOM5. Plant J. 2: 343 – 349

Buchanan-Wollaston V (1997) The molecular biology of leaf senescence. J. Exp. Bot. 48: 181 – 199

Buchanan-Wollaston V and Ainsworth C (1997) Leaf senescence in *Brassica napus:* cloning of senescence-related genes by subtractive hybridization. Plant Mol. Biol. 33: 821 – 834

Bufler G, Mor Y, Reid MS and Yang SF (1980) Changes in 1-aminocyclopropane-1-carboxylic acid content of cut carnation flowers in relation to their senescence. Planta 150: 439 – 442

Burg S and Burg EA (1965) Ethylene action and the ripening of fruits. Science 148: 1190 – 1148

Carey AT, Holt K, Picard S, Wilde R, Tucker GA, Bird CR, Schuch W and Seymour GB (1995) Tomato Exo-(1-4)-β-D-galactanase. Plant Physiol. 108: 1099 – 1107

Christoffersen RE, Warm E and Laties GG (1982) Gene expression during fruit ripening in avocado. Planta. 155: 52 – 57

Christoffersen RE, Tucker ML and Laties GG (1984) Cellulase gene expression in ripening avocado fruit: The accumulation of cellulase mRNA and protein as demonstrated by cDNA hybridization and immunodetection. Plant Mol. Biol. 3: 385 – 391

Cruz-Hernandez A and Gomez-Lim MA (1995) Alternative oxidase from mango (*Mangifera indica*, L.) is differentially regulated during fruit ripening. Planta. 197: 569 – 576

DellaPenna D, Alexander DC and Bennett AB (1986) Molecular cloning of tomato fruit polygalacturonase: Analysis of polygalacturonase mRNA levels during ripening. Proc. Natl. Acad. Sci. USA 83: 6420 – 6424

Denny FE (1924) Hastening the coloration of lemons. J. Agr. Res. 27: 757

De Vecchi L (1971) Fine structure of detached oat leaves senescing under different experimental conditions. Isr. J. Bot. 20: 169 – 183

Dong JG, Kim WT, Yip WK, Thompson GA, Li L, Bennett AB and Yang SF (1991) Cloning of a cDNA encoding 1-aminocyclopropane-1-carboxylate synthase and expression of its mRNA in ripening apple fruit. Planta. 185: 38 – 45

Dong JG, Fernandez-Maculet JC and Yang SF (1992) Purification and characterization of 1-aminocyclopropane-1-carboxylate oxidase from apple fruit. Proc. Natl. Acad. Sci. USA 89: 9789 – 9793

Dopico B, Lowe AL, Wilson ID, Merodio C and Grierson D (1993) Cloning and characterization of avocado fruit mRNAs and their expression during ripening and low-temperature storage. Plant Mol. Biol. 21: 437 – 449

Duncan RR, Bockholt AJ and Miller FR (1981) Descriptive comparison of senescent and nonsenescent sorghum genotypes. Agron. J. 73: 849 – 853

El-Antably HMM, Wareing PF and Hillman J (1967) Some physiological responses to D,L abscisin (dormin). Planta. 73: 74 – 90

Elliot KJ, Butler WO, Dickinson CD, Konno Y, Vedick TS, Fitzmaurice L and Mirkov TE (1993) Isolation and characterization of fruit vacuolar invertase genes from two tomato speices and temporal differences in mRNA levels during fruit ripening. Plant Mol. Biol. 21: 515 – 524

Ferrarese L, Trainotti L, Moretto P, de Laureto PP, Rascio N and Casadoro G (1995) Differential ethylene-inducible expression of cellulase in peppper plants. Plant Mol. Biol. 29: 735 – 747

Ferrie BJ, Beaudoin N, Burkhart W, Bowsher CG and Rothstein SJ (1994) The cloning of two tomato lipoxygenase genes and their differential expression during fruit ripening. Plant Physiol. 106: 109 – 118

Fils-Lycaon BR, Wiersma PA, Eastwell KC and Sautiere P (1996) A cherry protein and its gene, abundantly expressed in ripening fruit have been identified as thaumatin-like. Plant Physiol. 111: 269 – 273

Fischer RL and Bennett AB (1991) Role of cell wall hydrolases in fruit ripening. Ann. Rev. Plant Phys. Plant Mol. Biol. 42: 675 – 703

Fray RG, Lycett GW and Grierson D (1990) Nucleotide sequence of a heat-shock and ripening-related cDNA from tomato. Nuc. Acid Res. 23: 7148

Frenkel C, Klein I and Dilley DR (1968) Protein synthesis in relation to ripening of pome fruits. Plant Physiol. 43: 1146 – 1153

Gallego PP, Whotton L, Picton S, Grierson D and Gray JE (1995) A role for glutamate decarboxylase during tomato ripening: the characterisation of a cDNA encoding a putative glutamate decarboxylase with a calmodulin-binding site. Plant Mol. Biol. 27: 1143 – 1151

Gan S and Amasino RM (1995) Inhibition of leaf senescence by autoregulated production of cytokinin. Science 270: 1986 – 1988

Gane R (1934) Production of ethylene by some ripening fruit. Nature (London) 134: 1008

Garbarino JE, Oosumi T and Belknap WR (1995) Isolation of a polyubiquitin promoter and its expression in transgenic potato plants. Plant Physiol. 109: 1371 – 1378

Gilchrist D (1997) Mycotoxins reveal connections between plants and animals in apoptosis and ceramide signalling. Cell Death Differ. 4: 689 – 698

Giovannoni JJ, DellaPenna D, Bennett AB and Fischer RL (1989) Expression of a chimeric polygalacturonase gene in transgenic *rin* (ripening inhibitor) tomato fruit results in polyuronide degradation but not fruit softening. Plant Cell 1: 53 – 63

Grbic V and Bleecker AB (1995) Ethylene regulates the timing of leaf senescence in *Arabidopsis*. Plant J. 8: 595 – 602

Guzman P and Ecker JR (1990) Exploiting the triple response of *Arabidopsis* to identify ethylene-related mutants. Plant Cell 2: 513 – 523

Halevy AH, Whitehead CS and Kofranek AM (1984) Does pollination induce corolla abscission of cyclamen flowers by promoting ethylene production? Plant Physiol. 75: 1090 – 1093

Hall IV and Forsyth FR (1967) Production of ethylene by flowers following pollination and treatments with water and auxin. Can. J. Bot. 45: 1163 – 1166

Hall LN, Bird CR, Picton S, Tucker GA, Seymour GB and Grierson D (1994) Molecular characterisation of cDNA clones representing pectin esterase isozymes from tomato. Plant Mol. Biol. 25: 313 – 318

Hamilton AJ, Lycett GW and Grierson D (1990) Antisense gene that inhibits synthesis of the hormone ethylene in transgenic plants. Nature (London) 346: 284 – 287

Harpster MH, Lee KY and Dunsmuir P (1997) Isolation and characterization of a gene encoding endo-b-1,4-glucanase from pepper (*Capsicum annuum* L.). Plant Mol. Biol. 33: 47 – 59

Hensel LL, Grbic V, Baumgarten DA and Bleecker AB (1993) Developmental and age-related processes that influence the longevity and senescence of photosynthetic tissues in Arabidopsis. Plant Cell 5: 553 – 564

Hugueney P, Romer S, Kuntz M and Camara B (1992) Characterisation and molecular cloning of a flavoprotein catalyzing the synthesis of phytofluene and ζ-carotene in *Capsicum* chromoplasts. Eur. J. Biochem. 209: 399 – 407

John I, Drake R, Farrell A, Cooper W, Lee P, Horton P and Grierson D (1995) Delayed leaf senescence in ethylene-deficient ACC-oxidase antisense tomato plants: molecular and physiological analysis. Plant J. 7: 483 – 490

Kamachi K, Yamaya T, Hayakawa T, Mae T and Ojima K (1992) Changes in cytosolic glutamine synthetase polypeptide and its messenger RNA in a leaf blade of rice plants during natural senscence. Plant Physiol. 98: 1323 – 1329

Karvouni Z, John I, Taylor JE, Watson CF, Turner AJ and Grierson D (1995) Isolation and characterisation of a melon cDNA clone encoding phytoene synthase. Plant Mol. Biol. 27: 1153 – 1162

Kausch KD and Handa AK (1995) Molecular cloning and nucleotide sequence of a lipoxygenase cDNA from ripening tomato fruit. Plant Physiol. 107: 669 – 670

Kende H and Boller T (1981) Wound ethylene and 1-aminocyclopropane-1-carboxylate synthase in ripening tomato fruit. Planta. 151: 476 – 481

King GA, Davies KM, Stewart RJ and Borst WM (1995) Similarities in gene-expression during the post-harvest-induced senescence of spears and natural foliar senescence of asparagus. Plant Physiol. 108: 125 – 128

Klann E, Yelle S and Bennett AB (1992) Tomato fruit acid invertase complementary DNA. Plant Physiol. 99: 351 – 353

Kuntz M, Romer S, Suire C, Hugueney P, Weil JH, Schantz R and Camara B (1992) Identification of a cDNA for the plastid-located geranylgeranyl pyrophosphate synthase from *Capsicum annuum*; correlative increase in enzyme activity and transcript level during fruit ripening. Plant J. 2: 25 – 34

Kutsunai SY, Lin A-C, Percival FW, Laties GG and Christoffersen RE (1993) Ripening-related polygalacturonase cDNA from avocado. Plant Physiol. 103: 289 – 290

Lanahan MB, Yen H-C, Giovannoni JJ and Klee HJ (1994) The *Never Ripe* mutation blocks ethylene perception in tomato. Plant Cell 6: 521 – 530

Lashbrook CC, Gonzalez-Bosch C and Bennett AB (1994) Two divergent endo-β-1,4-glucanase genes exhibit overlapping expression in ripening fruit and abscising flowers. Plant Cell 6: 1485 – 1493

Lawton KA, Huang B, Goldsbrough PB and Woodson WR (1989) Molecular cloning and characterization of senescence-related genes from Carnation flower petals. Plant Physiol. 90: 690 – 696

Lawton KA, Raghothama KG, Goldsbrough PB and Woodson WR (1990) Regulation of senescence-related gene expression in carnation flower petals by ethylene. Plant Physiol. 93: 1370 – 1375

Lay-Yee M, Stead AD and Reid MS (1992) Flower senescence in daylily (*Hemerocallis*). Physiol. Plant 86: 308 – 314

Lay-Yee M and Knighton ML (1995) A full-length cDNA encoding 1-aminocyclopropane-1-carboxylate synthase from apple. Plant Physiol. 107: 1017 – 1018

Ledger SE and Gardner RC (1994) Cloning and characterization of five cDNAs for genes differentially expressed durign fruit development of kiwifruit (*Actinidia deliciosa* var. *deliciosa*). Plant Mol. Biol. 25: 877 – 886

Leopold AC (1961) Senescence in plant development. Science 134: 1727 – 1732

Leopold AC, Niedergang-Kamien E and Janick J (1959) Experimental modification of plant senescence. Plant Physiol. 34: 570 – 573

Lester DR, Speirs J, Orr G and Brady CJ (1994) Peach (*Prunus persica*) endopolygalacturonase cDNA isolation and mRNA analysis in melting and nonmelting peach cultivars. Plant Physiol. 105: 225 – 231

Li J, Nagpal P, Vitart V, McMorris TC and Chory J (1996) A role for brassinosteroids in light-dependent development of *Arabidopsis*. Science 272: 398 – 401

Lincoln JE, Cordes S, Read E and Fischer RL (1987) Regulation of gene expression by ethylene during *Lycopersicon esculentum* (tomato) fruit development. Proc. Natl. Acad. Sci. USA 84: 2793 – 2797

Lohman KN, Gan S, John MC and Amasino RM (1994) Molecular analysis of natural leaf senescence in *Arabidosis thaliana*. Physiol. Plant 92: 322 – 328

Longhurst T, Lee E, Hinde R, Brady C and Spiers J (1994) Structure of the tomato *Adh2* gene and *Adh2* pseudogenes, and a study of *Adh2* gene expression in fruit. Plant Mol. Biol. 26: 1073 – 1084

Lukaszewski TA and Reid MS (1989) Bulb-type flower senescence. Acta Hort. 261: 59 – 62

Makovetzki S and Goldschmidt EE (1976) A requirement for cytoplasmic protein synthesis during chloroplast senescence in the aquatic plant *Anarchis canadensis*. Plant Cell Physiol. 17: 859 – 862

Martin C and Thimann KV (1972) The role of protein synthesis in the senescence of leaves. I. The formation of protease. Plant Physiol. 49: 64 – 71

Martineau B, Houck CM, Sheehy RE and Hiatt WR (1994) Fruit-specific expression of the *A. tumefaciens* isopentenyl transferase gene in tomato: effects on fruit ripening and defense-related gene expression in leaves. Plant J. 5: 11 – 19

McGarvey DJ, Sirevag R and Christoffersen RE (1992) Ripening-related gene from avocado fruit. Plant Physiol. 98: 554 – 559

Meyer RC, Goldsbrough PB and Woodson WR (1991) An ethylene-responsive flower senescence-related gene from carnation encodes a protein homologous to glutathione S-transferases. Plant Mol. Biol. 17: 277 – 281

Meyer B, Houlne G, Pozueta-Romero J, Shantz ML and Schantz R (1996) Fruit-specific expression of a defensin-type gene family in bell pepper. Plant Physiol. 112: 615 – 622

Miki T, Yamamoto M, Nakagawa H, Ogura N, Mori H, Imaseki M and Sato T (1995) Nucleotide sequence of a cDNA for 1-aminocyclopropane-1-carboxylate synthase from melon fruits. Plant Physiol. 107: 297 – 298

Molisch H. 1938. *The Longevity of Plants* Lancaster, PA: Science Press

Morel J-B and Dangl JL (1997) The hypersensitive response and the induction of cell death in plants. Cell Death Differ. 4: 671 – 683

Mothes K and Engelbrecht L (1961) Kinetin-induced directed transport of substances in excised leaves in the dark. Phytochem. 1: 58 – 62

Nichols R (1966) Ethylene production during senescence of flowers. J. Hort. Sci. 41: 279 – 290

Nichols R (1968) The response of carnations (*Dianthus caryophyllus*) to ethylene. J. Hort. Sci. 43: 335 – 349

Nichols R, Bufler G, Mor Y, Fujino DW and Reid MS (1983) Changes in ethylene production and 1-aminocyclopropane-1-carboxylic acid content of pollinated carnation flowers. J. Plant Growth Regul. 2: 1 – 8

Oeller PW, Min-Wong L, Taylor LP, Pike DA and Theologis A (1991) Reversible inhibition of tomato fruit senescence by antisense RNA. Science 254: 437 – 439

O'Neill SD (1997) Pollination regulation of flower development. Ann. Rev. Plant Physiol. Mol. Biol. 48: 547 – 572

O'Neill SD and Nadeau J (1997) Postpollination flower development. Hortic. Rev. 19: 1 – 58

O'Neill SD, Nadeau JA, Zhang XS, Bui AQ and Halevy AH (1993) Interorgan regulation of ethylene biosynthetic genes by pollination. Plant Cell 5: 419 – 432

Osborne DJ (1962) Effect of kinetin on protein and nucleic acid metabolism in Xanthium leaves during senescence. Plant Physiol. 37: 595 – 602

Payton S, Fray RG, Brown S and Grierson D (1996) Ethylene receptor expression is regulated during fruit ripening, flower senescence and abscission. Plant Mol. Biol. 31: 1227 – 1231

Pear JR, Ridge N, Rasmussen R, Rose RE and Houck CM (1989) Isolation and characterization of a fruit specific cDNA and the corresponding genomic clone from tomato. Plant Mol. Biol. 13: 639 – 651

Pearl R and Miner JR (1935) Experimental studies on the duration of life. XIV. The comparative mortality of certain lower organisms. Q. Rev. Biol. 10: 60 – 79

Pecker I, Chamovitz D, Linden H, Sandmann G and Hirschberg J (1992) A single polypeptide catalyzing the conversion of phytoene to ζ-carotene is transcriptionally regulated during tomato fruit ripening. Proc. Natl. Acad. Sci. USA 89: 4962 – 4966

Picton S, Barton SL, Bouzayen M, Hamilton AJ and Grierson D (1993a) Altered fruit ripening and leaf senescence in tomatoes expressing an antisense ethylene-forming enzyme transgene. Plant J. 3: 469 – 481

Picton S, Gray J, Barton S, AbuBakar U, Lowe A and Grierson D (1993b) cDNA cloning and characterisation of novel ripening-related mRNAs with altered patterns of accumulation in the *ripening inhibitor* (*rin*) tomato ripening mutant. Plant Mol. Biol. 23: 193 – 207

Pierce RO, Knowles PF and Phillips DA (1984) Inheritance of delayed leaf senescence in Soybean. Crop Sci. 24: 515 – 517

Porat R, Borochov A, Halevy AH and O'Neill SD (1994) Pollination-induced senescence of Phalaenopsis petals – The wilting process, ethylene production and sensitivity to ethylene. Plant Growth Reg. 15: 129 – 136

Pozueta-Romero J, Klein M, Houlne G, Schantz M, Meyer B and Schantz R (1995) Characterization of a family of genes encoding a fruit-specific wound-stimulated protein of bell pepper (*Capsicum annuum*): identification of a new family of transposable elements. Plant Mol. Biol. 28: 1011 – 1025

Rastogi R, Dulson J and Rothstein SJ (1993) Cloning of tomato (*Lycopersicon esculentum* Mill.) arginine decarboxylase gene and its expression during fruit ripening. Plant Physiol. 103: 829 – 834

Richmond AE and Lang A (1957) Effect of kinetin on protein content and survival of detached Xanthium leaves. Science 125: 650 – 651

Romer S, Hugueney P, Bouvier F, Camara B and Kuntz M (1993) Expression of the genes encoding the early carotenoid biosynthetic enzymes in *Capsicum annuum*. Biochem. Biophys. Res. Comm. 196: 1414 – 1421

Rose JKC, Lee HH and Bennett AB (1997) Expression of a divergent expansin gene is fruit-specific and ripening-regulated. Proc. Natl. Acad. Sci. USA 94:5955 – 5960

Ross GS, Knighton ML and Lay-Yee M (1992) An ethylene-related cDNA from ripening apples. Plant Mol. Biol. 19: 231 – 238

Ross GS, Wegrzyn T, MacRae EA and Redgwell RJ (1994) Apple β-galactosidase. Plant Physiol. 106: 521 – 528

Sachs J. 1882. Text-Book of Botany. 2 ed. Oxford: Clarendon Press

Sato T and Theologis A (1989) Cloning the mRNA encoding 1-aminocyclopropane-1-carboxylate synthase, the key enzyme for ethylene biosynthesis in plants. Proc. Natl. Acad. Sci. USA 86: 6621 – 6625

Sheehy RE, Pearson J, Brady CJ and Hiatt WR (1987) Molecular characterization of tomato fruit polygalacturonase. Mol. Gen. Genet. 208: 30 – 36

Sheehy RE, Kramer M and Hiatt WR (1988) Reduction of polygalacturonase activity in tomato fruit by antisense RNA. Proc. Natl. Acad. Sci. USA 85: 8805 – 8809

Shibaoka H and Thimann KV (1970) Antagonism between kinetin and amino acids: experiments on the mode of action of cytokinin. Plant Physiol. 46: 212 – 220

Sievers AF and True RH (1912) A preliminary study of the forced curing of lemons as practiced in California. US Dept. AG. B. Plant. Ind. Bull. 232

Skadsen RW and Cherry JH (1983) Quantitative changes in *in vitro* and *in vivo* protein synthesis in aging and rejuvenated soybean cotyledons. Plant Physiol. 71: 861 – 868

Slater A, Maunders MJ, Edwards K, Schuch W and Grierson D (1985) Isolation and characterization of cDNA clones for tomato polygalacturonase and other ripening related proteins. Plant Mol. Biol. 5: 137 – 147

Smart CM, Hosken SE, Thomas H, Greaves JA, Blair BG and Schuch W (1995) The timing of maize leaf senescence and characterization of senscence-related cDNAs. Physiol. Plant 93: 673 – 682

Smith CJS, Watson CF, Ray J, Bird CR, Morris PC, Schuch W and Grierson D (1988) Antisense RNA inhibition of polygalacturonase gene expression in transgenic tomatoes. Nature (London) 334: 724 – 726

Speirs J and Brady CJ (1981) A co-ordinated decline in the synthesis of subunits of ribulosebisphosphate carboxylase in aging wheat leaves. II. Abundance of messenger RNA. Aust. J. Plant Physiol. 8: 603 – 618

Speirs J, Brady CJ, Grierson D and Lee E (1984) Changes in ribosome organization and messenger RNA abundance in ripening tomato fruits. Aust. J. Plant Physiol. 11: 225 – 233

Sylvestre I, Droillard M-J, Bureau J-M and Paulin A (1989) Effects of the ethylene rise on the peroxidation of membrane lipids during the senescence of cut carnations. Plant Physiol. Biochem. 27: 407 – 413

Taylor CB, Bariloa PA, Delcardayre SB, Raines RT and Green PJ (1993) RNS2-a senescence-associated RNase of *Arabidopsis* that diverged from the S-RNAses before speciation. Proc. Natl. Acad. Sci. USA 90: 5118 – 5122

Thomas H (1976) Delayed senescence in leaves treated with the protein synthesis inhibitor MDMP. Plant Sci. Lett. 6: 369 – 377

Thomas H (1987) *Sid*: a Mendelian locus controlling thylakoid membrane disassembly in senescing leaves of *Festuca pratensis*. Theor. Appl. Genet. 73: 551 – 555

Thomas H and Stoddart JL (1975) Separation of chlorophyll degradation from other senescence processes in leaves of a mutant genotype of meadow fesuce (*Festuca pratensis*). Plant Physiol. 56: 438 – 441

Thomas H and Stoddart JL (1980) Leaf Senescence. Ann. Rev. Plant Physiol. 31: 83 – 111

Tigchelaar EC, McGlasson WB and Buescher RW (1978) Genetic regulation of tomato fruit ripening. HortSci. 13: 508 – 513

Turner LA, Kausch KD and Handa AK (1996) Isolation and nucleotide sequence of two cDNAs corresponding to tomato fruit pectin methylesterase genes. Plant Physiol. 111: 652

Ueda J and Kato J (1980) Isolation and identification of a senescence-promoting substance from wormwood (*Artemisia absinthuim* L.). Plant Physiol. 66: 246 – 249

Van Der Straeten D, Van Wiemeersch L, Goodman HM and Van Montagu M (1990) Cloning and sequence of two different cDNAs encoding 1-aminocyclopropane-1-carboxylate synthase in tomato. Proc. Natl.Acad. Sci. USA 87: 4859 – 4863

Whitehead CS, Fujino DW and Reid MS (1983) Identification of the ethylene precursor, 1-aminocyclopropane-1-carboxylic acid (ACC) in pollen. Scienti. Hort. 21: 291 – 297

Whittaker DJ, Smith CS and Gardner RC (1995) Three cDNAs encoding S-adenosyl-L-methionine synthetase from *Actinidia chinensis*. Plant Physiol. 108: 1307 – 1308

Wilkinson JQ, Lanahan MB, Conner TW and Klee HJ (1995a) Identification of mRNAs with enhanced expression in ripening strawberry fruit using polymerase chain reaction differential display. Plant Mol Biol 27: 1097 – 1108

Wilkinson JQ, Lanahan MB, Yen HC, Giovannoni JJ and Klee JH (1995b) An ethylene-inducible component of signal transduction encoded by *Never-ripe*. Science 1807 – 1809

Woodson WR (1987) Changes in protein and mRNA populations during the senescence of carnation petals. Physiol Plant 71: 495 – 502

Yu SM and Kao CH (1981) Retardation of leaf senescence by inhibitors of RNA and protein synthesis. Physiol Plant 52: 207 – 210