



## Review

## Ethylene control in cut flowers: Classical and innovative approaches

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## ABSTRACT

Ethylene-mediated premature floral senescence and petal or flower abscission affect postharvest longevity of several species used as cut flowers. Exposure to exogenous or endogenously produced ethylene can be controlled in several ways. These include the use of ethylene biosynthesis inhibitors or ethylene action inhibitors, and ethylene removal technologies. In addition, genetic modification can be very effective in controlling ethylene synthesis and perception. We review here the potential for applications of nanotechnology to control ethylene levels and postharvest management in the flower industry. Already, nanosponges have been shown to enhance efficacy of the ethylene inhibitor 1-MCP in several flower species. In carnation, 1-MCP included in nanosponges also allowed better control of *Botrytis cinerea* damage. However other applications are also considered based on successes in the use of this technology to increase agricultural production and decrease postharvest waste. Nano-metal based sensors could be used for detection of ethylene in the store and to label the product along the distribution chain. Furthermore, nanocomposites could be included as scavengers for ethylene removal in active packaging, and nanocatalysts could promote ethylene catalytic degradation in the warehouse. Nanoparticles could also be introduced into a new generation of packaging to control effects of gases and UV, and increase strength, quality and packaging appearance. This review highlights recent results on the use of nanotechnology *sensu lato* and potential application for cut flower vase life improvement, focusing on ethylene control strategies.

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## 1. Introduction

Postharvest performance is a key factor in the commercial value of cut flowers. Although external quality criteria such as appearance, colour and uniformity, are the major variables that influence the consumer's decision to purchase cut flowers, their longevity is fundamental to convince the consumer to re-purchase them (Reid and Jjiang, 2012). As a fresh commodity and because of their extreme sensitivity, cut flowers are vulnerable to large postharvest losses. In addition to developmental senescence, cut flowers are also subject to leaf discoloration, premature wilting, and disease from moulds and fungal pathogens. An integrated approach is therefore adopted to maintain quality throughout the distribution chain to reduce water loss (e.g. avoiding high temperatures), control disease (such

as *Botrytis* and *Alternaria*) and to limit cut flower ageing (avoiding prolonged cold storage).

Advances in postharvest science and technology aim to provide information for the horticultural industry to enable them to supply attractive and long-lived flowers to consumers. Indeed in the last ten years, substantial progress in postharvest technologies has been achieved including novel packaging, storage and transport systems, pest and disease control for market access, senescence control, supply chain optimization, and track and trace systems to ensure delivery of premium quality products to markets (Toivonen, 2007; Michailides and Manganaris, 2009; Sharma and Alemwati, 2010). Chemicals are used extensively in modern agriculture in order to improve yield and quality. However, their use poses environmental and public health concerns. Many chemicals that affect ethylene synthesis or its action, which are currently in use to extend the shelf-life of flowers, may be soon banned due to their environmental impact. Over the last decades, environmentally and health-friendly production methods and conscientious use of resources have become crucial for reaching the goal of more sustainable plant production. Thus further progress will require an

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integration of available bio-, info- and nano-technologies through a systems biology approach.

### 1.1. Role of ethylene in floral senescence

Ethylene is a simple molecule composed of two carbon atoms symmetrically linked by a double bond and it naturally occurs in gaseous form. It is, furthermore, a plant growth regulator involved in the regulation of a wide range of different physiological processes, including germination, growth, floral initiation and opening, both leaf and floral senescence as well as organ abscission and fruit ripening (Yoo et al., 2009).

#### 1.1.1. Ethylene as an endogenous and exogenous regulator

Floral lifespan is often terminated by the abscission of petals that are still turgid, or by petal wilting or withering. In many species, these processes are regulated by ethylene (van Doorn, 2001; van Doorn and Woltering, 2008) through changes in endogenous levels. Plant tissues synthesize small amounts of ethylene ( $0.1\text{--}0.2\ \mu\text{L kg}^{-1}\ \text{h}^{-1}$ ; Martínez-Romero et al., 2007). However ethylene production changes during plant development and in relation to physiological status (Yang and Hoffman, 1984).

In many species exogenous ethylene can also accelerate floral senescence. Ethylene is produced by many plant tissues (Gane, 1934) and other sources, including bacterial and fungal fermentation processes, and pyrolysis of hydrocarbons, which releases ethylene as a component of air pollutants (Cape, 2003), all of which can thus affect the longevity of cut flowers in the horticultural supply chain. Ethylene is biologically active at very low concentrations ( $\text{nL--}\mu\text{L L}^{-1}$ ), but there are significant differences in ethylene sensitivity between species and even cultivars of the same species (Serek et al., 2006b; Scariot et al., 2008). A detailed classification of flowers based on ethylene sensitivity is reported by van Doorn (2001).

#### 1.1.2. Plant species: sensitivity and effects

Responses to ethylene vary widely according to the species (Reid and Wu, 1992) although they are often consistent within either families or subfamilies (van Doorn, 2001). Ethylene-sensitive species include a number of important cut flowers. For example petals of orchids (*Phalaenopsis*), *Hibiscus* (Çelikel and Reid, 2002), and carnation (*Diathus caryophyllus*) (Serek et al., 1995a,b) wilt in response to ethylene. In other species, such as *Antirrhinum majus*, *Rosa hybrida* (Serek et al., 1995a), and wax flower (*Chamelacium uncinatum*) (Macnish et al., 2000), ethylene induces petal or flower abscission.

Ethylene sensitive flowers can be classified into three types (Kumar et al., 2008). First, those like carnation and petunia where senescence is regulated by an increased amount of ethylene production either with ageing or following pollination (Serek et al., 1995a). Second, for example cyclamen, which only become sensitive to ethylene and produce increased amounts of the hormone when they are pollinated (Halevy et al., 1984). Third, such as rose, which are sensitive to ethylene upon flower bud opening but do not produce elevated amounts of ethylene as they age (Kumar et al., 2008).

As well as accelerating petal senescence and deterioration, ethylene (either endogenous or from an external source) can induce other undesirable physiological disorders to vegetative and flowering organs during postharvest storage of cut flowers both in monocotyledons and dicotyledons including pathogen susceptibility (McKenzie and Lovell, 1992; van Doorn, 2001). For example, *Botrytis cinerea* is one of the most significant postharvest fungal pathogens causing losses in ornamental plants. Disease caused by this fungus has been shown to be enhanced by the presence of ethylene in rose and carnation (Elad, 1988; Seglie et al., 2012). However, depending on the type of pathogen and plant species, the role

of ethylene can be dramatically different. Indeed plants deficient in ethylene signalling may show either increased susceptibility or increased resistance (Elad, 1988).

Thus data on ethylene sensitivity of cut flower species is important for predicting effects of exposure during the supply chain such as mixed storage and transport of flowers with fruit species. It is also needed to evaluate the appropriateness of treatments to reduce ethylene production or exposure and to inform breeding programmes aimed at improving flower vase life.

### 1.2. Ethylene control strategies

Ethylene biosynthesis, perception, and signal transduction are well-documented as well as its regulation at biochemical and genetic levels (reviewed in Wang et al., 2002). This knowledge has been used to develop different strategies to reduce ethylene production or inhibit its action (either with new cultivars or vase-life treatments), and in turn to prolong flower postharvest performance.

Premature senescence and abscission caused by exposure to exogenous or endogenous ethylene can be mitigated in several ways (Fig. 1) including ethylene biosynthesis inhibitors, ethylene action inhibitors and ethylene removal technologies (reviewed in Martínez-Romero et al., 2007). Genetic modification is also a very effective way of controlling ethylene synthesis and perception. Attempts to obtain plants with both reduced endogenous ethylene biosynthesis or a reduced ethylene sensitivity have been reviewed by Serek et al. (2006b).

#### 1.2.1. Genetic strategies

Changes in gene expression during petal senescence have been studied through transcriptomics of a number of model flowers (e.g. *Petunia*, *Arabidopsis*) and cut flower species (e.g. *Alstroemeria*, *Dianthus*, *Iris*, *Sandersonia*) (Rogers, 2013). In species where petal senescence is ethylene-sensitive, groups of genes can be identified that are ethylene-regulated, comprising transcription factors, genes encoding for enzymes in the biosynthetic pathway for ethylene production, ethylene receptors and ethylene signalling and responsive genes (Rogers, 2013).

Ethylene biosynthesis is primarily regulated by 1-aminocyclopropane-1-carboxylic acid (ACC) synthase (ACS) and ACC oxidase (ACO) and an early success by the company Florigene in delaying carnation floral senescence was through antisense down-regulation of ACO (Savin et al., 1995). This success was closely followed by down-regulation of ACO in other flower species such as begonia (Einset and Kopperud, 1995) and torenia (Aida et al., 1998). Down-regulation of the ACS gene in carnation also reduced ethylene production (Kiss et al., 2000). Use of antisense sequences in petunia for ACO and ACS, derived heterologously from broccoli, also delayed floral senescence (Huang et al., 2007), showing that the approach can be used more broadly. However, these strategies have no effect when flowers are exposed to exogenous ethylene, as can occur during transit and marketing.

A more effective approach to protecting flowers from exogenous ethylene in the supply chain is therefore to focus on ethylene perception. Ethylene perception occurs through a well-conserved signalling pathway and the receptor is encoded by a family of five genes: *ETR1*, *ETR2*, *EIN4*, *ERS1* and *ERS2* (Yoo et al., 2009). Again an early discovery was that expression of a mutated *ETR1* gene from *Arabidopsis* (*etr1-1*) disrupts ethylene signalling in a wide range of heterologous species (Bleecker et al., 1988; Wilkinson et al., 1997), making it an extremely useful tool (Binder, 2008; Serek et al., 2006a). It has been used successfully in a range of ornamental species to delay floral senescence including *Petunia* (Clevenger et al., 2004; Clark et al., 1999a; Gubrium et al., 2000;

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