



## **Literature Review**

# **1-Methylcyclopropene: a review of its use on potato tubers**

**Report Author: G. Harper, Sutton Bridge Crop  
Storage Research**

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## 1. Summary

Ethylene decreases sprout elongation in all varieties and with commercial acceptability for some varieties. However its use may be associated with unacceptable fry colours. One strategy being pursued to counteract this adverse effect of ethylene is an additional treatment with 1-methylcyclopropene (1-MCP), marketed as SmartFresh™. 1-MCP is an inhibitor of ethylene perception that can markedly reduce the ripening and senescence processes of many horticultural commodities. This review examines the current state of knowledge of the use of 1-MCP on potato

The response to ethylene is variety specific. Using commercial criteria, the fry colour of tubers stored under 10ppm ethylene at 9°C is generally commercially acceptable with a few exceptions. These exceptions include crisping varieties. 1-MCP does decrease ethylene affected sugar levels and fry colour in some varieties. However, because of the relatively small observed effects of ethylene on fry colour of tubers stored at 9°C, these decreases are small.

There are differences in varietal response to 1-MCP which must be empirically determined and further optimisation of the timing and concentration of treatment would be of particular benefit to varieties found responsive. The cost-benefit of 1-MCP should be individually established for each variety for which ethylene is used.

## 2. Introduction

Potato tubers may be stored for greater periods than their natural dormancy, to supply the processing industry with raw material year round. Once tuber dormancy is broken, sprouting occurs with loss in quality due both to disease-related and physiological processes (Burton *et al.*, 1992). To extend the storage time for potato tubers beyond that of natural dormancy (generally 1-15 weeks) two main strategies are employed by the potato processing industry: storage at low temperatures (expensive and effects quality); and by the use of chemical sprout suppressants such as CIPC. Alternative approaches to sprout suppression are at various stages of development and commercialisation. For example, the continuous application of ethylene (4 ppm) to potatoes during storage reduces the length of sprouts once they have initiated (Prange *et al.*, 1998). Ethylene has hence been used as an alternative to CIPC in the UK fresh market potato sector.

Although continuous exposure to ethylene gas controls sprout growth, for some cultivars it can also result in darker fry colour. Additionally variation in sprout control between varieties can be reduced by lowering the storage temperature below 6°C although this often stimulates low-temperature sweetening, leading to the accumulation of reducing-sugars (fructose and glucose) (reviewed in Sowokinos, 2001). One strategy being developed to counteract the adverse effects of ethylene is treatment with 1-methylcyclopropene (1-MCP), marketed as SmartFresh™. 1-MCP is an inhibitor of ethylene perception that can markedly reduce the ripening and senescence processes of many horticultural commodities (Watkins 2006).

This review of the use of 1-MCP for stored potato aims to describe the factors affecting the efficacy of the treatment, how these factors have been taken account of in past potato storage research and to identify future strategies to maximise the potential use of this treatment.

### **3. Background to use of 1-MCP**

#### ***Mode of action***

Ethylene, a gas, is a plant hormone that plays multiple roles in regulating plant growth and development and is a key modulator of the responses to biotic or abiotic stresses. Ethylene is perceived by membrane-located receptor proteins including those encoded by ETR (Ethylene Response) and ERS (Ethylene Response Sensor) genes. In all plants examined to date, the ethylene receptors exist as a multi-member family (reviewed in Binder *et al.*, 2012). Receptor binding triggers a downstream signalling cascade (reviewed in Klee and Tieman, 2002; Binder, 2008). In the absence of ethylene the receptors act as negative regulators in the ethylene-signalling pathway by their activation of CTR1 (Constitutive Triple Response 1) a Serine/Threonine kinase that suppresses the ethylene response (Hua and Meyerowitz, 1998). More recent data indicates that, in addition to their general role in ethylene perception, receptors have more individual or specific roles in transducing the ethylene signal (reviewed Shakeel *et al.*, 2013).

1-MCP binds to ethylene receptors with some 10-fold greater affinity than ethylene (reviewed Blankenship and Dole 2003) and such receptor occupation prevents the binding of endogenous and exogenous ethylene, and the concomitant effect(s) of this hormone (Sisler and Serek 1997; Binder and Bleecker 2003). 1-MCP

also affects ethylene biosynthesis, in at least some species, through the inhibition of positive feedback mechanisms that stimulate autocatalytic ethylene production in climacteric fruits. 1-MCP can inhibit the activities of enzymes involved in ethylene biosynthesis so reducing ethylene levels. Further, 1-MCP decreases gene expression of these enzymes and of ethylene receptors (Ma *et al.*, 2009). Plants remain insensitive to ethylene for as long as 1-MCP is bound to the receptor (Sisler, 2006). The competitive inhibition of the ethylene receptor complex by 1-MCP suggests there is a continual competition between ethylene and 1-MCP for receptor binding sites. Moreover, turnover and synthesis of new receptors can require additional treatment with 1-MCP to ensure complete protection from the effects of ethylene. Differences in ethylene and 1-MCP concentration, treatment duration, timing, and temperature and other factors may result in differential gene expression, since physiological and biochemical responses differ depending on these parameters (Blankenship and Dole, 2003; Watkins, 2006; Bufler, 2009). The actions and effects of ethylene on potato were reviewed by Briddon (2006)

### ***Commercial use of 1-MCP***

A patent on the use of cyclopropenes to inhibit ethylene action was granted in 1996 to Edward Sisler and Sylvia Blankenship, North Carolina State University, USA. Further development by others produced a formulation that releases 1-MCP when mixed with water. AgroFresh, a subsidiary of Dow Chemical Company, are commercial rights holders for use of 1-MCP on edible crops under the trade name SmartFresh™. 1-MCP is apparently non-toxic at active concentrations and, as SmartFresh™, complies with FAO specifications. There are no significant limits to its use in commercial environments. Toxicity tests show that 1-MCP is not expected to be harmful to living organisms or the environment. At standard temperature and pressure 1-MCP is a gas that decays, dissipates or binds to ethylene receptors such that there is no or negligible residue within 24 hours of application. 1-MCP is formulated as a gas encapsulated with  $\gamma$ -cyclodextrin; 1-MCP is released on the addition of water.

1-MCP has been approved and accepted for use on a wide range of agricultural commodities including, cut flowers, apples, tomatoes, kiwis, bananas and melons in many countries including the European Union and the United States.

## ***Method of treatment***

1-MCP is combined with other materials for handling and mixed with a specific amount of water or other solution to release it as a gas. The commercial formulation for application is propriety information of Dow Chemical Company. It is used within enclosed sites to maintain an effective working concentration for the duration of treatment.

## **4. Factors Influencing efficacy of 1-MCP**

### ***Concentration of 1-MCP treatment.***

The affinity of 1-MCP for ethylene receptors is about 10 times greater than that of ethylene hence 1-MCP is active at much lower concentrations than ethylene. However, the effective concentrations of 1-MCP vary widely with commodity and with factors that affect physiological status. These include maturity and crop condition, and timing, duration, temperature and method of application (reviewed Blankenship and Dole, 2003). The review recorded the 1-MCP concentrations trialled on a wide range of commodities. The effective concentration varied over ~5 orders of magnitude. 2.5 nl l<sup>-1</sup> induced senescence of *Dianthus caryophyllus* (Carnation, Sisler *et al.*, 1996a), 100 nl l<sup>-1</sup> applied to *Citrus* spp. (orange fruit) blocked ethylene induced degreening and inhibited abscission (Porat *et al.*, 1999). In apples, 1 µl l<sup>-1</sup> was required to block ethylene action (Jiang and Joyce, 2002) and 12 µl l<sup>-1</sup> were effective in blocking ethylene action in broccoli. (Able *et al.*, 2002a). Hofman *et al.* (2001) reported that 100 µl l<sup>-1</sup> 1-MCP was required to increase shelf life in *Mangifera indica* (mango).

The maximum allowable concentration of 1-MCP that can currently be applied in the UK is 1000 ppb. Published trial results on stored potato under ethylene have used 1-MCP at a concentration of 0.9 µL.L<sup>-1</sup> (900 ppb, Prange *et al.*, 2005, Daniels-Lake *et al.*, 2008). In Potato Council-funded studies (R441, Defra LK09127/R464) a concentration of 625 ppb 1-MCP was used. This was a concentration used for other long term stored commodities, for example in the apple storage industry this is an effective concentration and provides an economic benefit. In another study (R464), the effect of 325, 625 and 1000 ppb 1-MCP treatment prior to 10ppm ethylene storage on fry colour and sugar concentration was studied. The lightest fry colours were generally observed at the highest 1-MCP treatment concentration. Statistically significant differences in fry colour for Desiree were found between with and without 1-

MCP treatment but not between the different treatment concentrations. There were no statistically significant differences for Saturna or Markies between with and without 1-MCP treatment at either sampling occasion. Increasing the concentration 1-MCP treatment from 390 to 1000 ppb decreased reducing sugar concentration after both 2 and 4 months storage in Markies, although the effect in Russet Burbank and Saturna was much less marked.

### ***Duration of 1-MCP treatment***

Most trials have used a treatment duration of between 12 to 24 h - which was sufficient to achieve a full response. Generally shorter exposure times were considered where crop was treated at ambient temperatures, while 24 hours was considered ample for commodities treated at lower temperatures (1-5°C). Short exposures may not be sufficient as a 6 h exposure to 1-MCP was not enough to induce respiratory or ethylene production changes in avocado (Jeong *et al.*, 2002). There is some evidence that different varieties of a commodity may require different treatments. For example 'Empire' apples required less treatment time than 'Cortland' to achieve the same effect at the same 1-MCP concentration (DeEll *et al.*, 2002). A standard duration of 24 h exposure has been used in all Potato Council trials of 1-MCP.

### ***Plant developmental stage, plant maturity and varietal effects***

The effects of 1-MCP vary with plant maturity. The effect of 1-MCP in apricots (*Prunus armeniaca*) decreased as fruit development increased (Fan *et al.*, 2000a). Leafy brassica was found to respond less to 1-MCP than a floral brassica, thought to be due to ages of leaves and inherent differences between flowers and leaves (Able *et al.*, 2002a). Ripeness in pears was found to have significant 1-MCP treatment effects. When testing pears for tissue mechanical properties (Baritelle *et al.*, 2001), treatment of under ripe pears can lead to a loss of ripening capacity on removal from storage. In the Redchief strain of 'Delicious' apples advancing maturity slightly decreased the effect of 1-MCP (Mir *et al.*, 2001).

There have been no studies investigating the maturity of potato crops in relation to 1-MCP response. Variability of response to 1-MCP between potato varieties has been previously recorded (Daniels-Lake, 2008; Coleman, P., Greenvale AP, pers.

comm.). Variation included both the strength and duration of the protective effect of 1-MCP (Daniels-Lake *et al.*, unpublished data). Variability was also observed in more recent trials of GB varieties (Foukaraki 2013, Colgan *et al.*, 2013). Colgan *et al.* (2013) found little effect of ethylene or of 1-MCP, singly or in combination, on sugar levels in Hermes, Russet Burbank or Saturna but a protective effect of 1-MCP was observed in Cabaret and Sylvana.

Foukaraki (2013) suggested that the inability of 1-MCP to block the effect of ethylene in certain cultivars was associated with skin thickness, with higher thickness a barrier for 1-MCP to reach ethylene binding sites. An alternative explanation is that it is related to differences in tissue density between varieties (R. Colgan pers. comm.).

### ***Timing of 1-MCP treatment***

Although the importance of time from harvest to 1-MCP treatment varies with the crop species, earlier rather than later applications are consistently more successful in preserving quality. For example, 1-MCP must be applied as soon as possible to broccoli and pak choy (*Brassica rapa*, Able *et al.*, 2002a). Ethylene production, softening, and internal browning in ripening apricots and plums was inhibited when fruit were treated with 1-MCP after storage, but not before storage (Dong *et al.*, 2002). 1-MCP prevented some of the consequences of exogenous ethylene when the two compounds were applied together but completely prevented ethylene damage when the 1-MCP was applied prior to the ethylene. This indicates that 1-MCP ideally should be applied prior to any possible ethylene exposures (Serek *et al.*, 1995c).

In potato a 24h 1-MCP application prior to storage of tubers in ethylene successfully suppressed the action of ethylene in terms of sugar accumulation in a variety dependent manner. Application of 1-MCP at 900 ppb 24 hours prior to storage of tubers in 10 ppm ethylene at 6°C significantly lowered fructose, glucose and sucrose concentrations at six weeks after time of first indication of sprouting and after 26 weeks storage in Marfona but not in Estima (Foukaraki *et al.*, 2011; Foukaraki 2013). Additionally Foukaraki (2013) found that 1-MCP treatment prior to storage of tubers in 10 ppm ethylene at 6°C was more effective than when applied at the time of first indication of sprouting in suppressing some actions of ethylene. In studies of Colgan *et al.* (2013) 1-MCP (625 ppb) was applied prior to storage of tubers at 9°C

under 10 ppm ethylene. This also successfully suppressed the action of ethylene in terms of sugar accumulation in a cultivar dependent manner.

### ***Frequency of 1-MCP application***

The effect of repeat applications of 1-MCP varies with commodity, crop physiological condition and variety. Two applications were found to be more beneficial in reducing ethylene-induced mesocarp discoloration in avocado than just one application (Pesis *et al.*, 2002) whereas multiple applications of 1-MCP to broccoli and pak choy had no more experimental effect than a single application (Able *et al.*, 2002a). Repeated 1-MCP applications can further reduce ripening of pear (Ekman *et al.*, 2004),

The optimal timing of 1-MCP treatment in relation to stored potato is still relatively undefined. Typically 1-MCP has been applied prior to ethylene treatment. Prange *et al.* (2005) applied 1-MCP at least 12 weeks (Shepody) and eight weeks (Russet Burbank) after harvest and with monthly and bi-monthly additional treatments. A single initial dose was effective to control increase in fry colour for Russet Burbank whereas Shepody required additional 1-MCP treatment for adequate fry colour control. Daniels-Lake *et al.* (2008) applied 1-MCP to Russet Burbank following an eight week curing and pull down period and, as with Prange *et al.* (2008) found a single treatment effective for this variety.

In Potato Council-supported trials (Colgan *et al.*, 2013; R412; R464) 1-MCP was applied as soon as possible after receipt, and usually within 1 week of harvest. Additional applications were made at bi-monthly intervals (Colgan *et al.*, 2013) or at a defined point during emergence from dormancy break (R412). The results from these studies suggest that that a single initial 1-MCP application made as soon as possible following harvest was effective in reducing fry colours in responsive varieties. The effects of subsequent treatments, at two monthly intervals, were variety dependent. For example with the varieties Cabaret and Sylvana single and multiple applications of 1-MCP were able to suppress the accumulation of reducing sugars during the first 4 months of storage but there was no such effect in Russet Burbank (Colgan *et al.* 2013).

## ***Effects of 1-MCP in the presence of endogenous or exogenous ethylene***

Studies of 1-MCP have been conducted in the presence of applied ethylene and/or in the presence of endogenous ethylene. Responses to the two types of tests vary by commodity. Some crops will benefit from 1-MCP regardless of the presence of exogenous ethylene. Other crops show little benefit from 1-MCP application unless exogenous ethylene is present. If exogenous ethylene was present, 1-MCP protected both broccoli and pak choy from detrimental ethylene effects. In contrast, in the absence of exogenous ethylene, 1-MCP had little effect in extending shelf life of pak choy but did extend the shelf life of broccoli by 20% (Able *et al.*, 2002a). When strawberry fruits were treated with 1-MCP and then exposed to an exogenous application of ethylene, the effects were dependent on fruit maturity and 1-MCP concentration (Ku *et al.*, 1999; Tian *et al.*, 2000). However, 1-MCP was also found to extend strawberry postharvest life in the absence of exogenous ethylene (Jiang *et al.*, 2001).

Exposure of potato tubers to 1-MCP did not suppress the action of ethylene in increasing the respiration rate or ethylene production at different stages during storage of potatoes (Foukaraki, 2013)

## ***Regeneration of ethylene receptors***

An assumption has been made that 1-MCP permanently binds to receptors present at the time of treatment and subsequent development of ethylene sensitivity is due to appearance of new sites. Receptor regeneration may provide an explanation for some differences in the effect of various ethylene receptor blockers (Cameron and Reid 2001).

In avocado, ripening was delayed by about 2 weeks following 1-MCP treatment, fruit then ripened normally (Feng *et al.*, 2000). Pesis *et al.* (2002) found that two treatments of 300 nl l<sup>-1</sup> applied 10 days apart, was enough to prevent softening in avocado, whereas a single application allowed fruit to soften normally. Able *et al.* (2002a) suggested that ethylene binding sites are regenerated after treatment of broccoli as continuous application was more effective than a single treatment. However, this was not true in pak choy. Ripening was delayed in tomatoes by 5–10 days with one application of 1-MCP but fruit must be retreated for continued effect

(Hoeberichts *et al.*, 2002; Wills and Ku, 2002; Sisler *et al.*, 1996b). Partial responses in some apple cultivars either seem to suggest these cultivars are able to regenerate sites or that binding is incomplete. 'McIntosh' apples, for instance, may need higher 1-MCP concentrations, perhaps because this cultivar produces large amounts of ethylene (Watkins *et al.*, 2000).

In potato, Prange *et al.* (2005) suggested the production of new ethylene-binding sites. Kools *et al.* (2012) proposed that ethylene and 1-MCP bind with different affinities to different ethylene receptors in onion.

### ***Factors affecting binding of 1-MCP to ethylene receptors***

In Russet Burbank potato tubers, 1-MCP has been used to reduce the reported detrimental effect of ethylene on fry color darkening. 1-MCP did not interfere with ethylene-induced sprout suppression, and ethylene did not cause such a dark fry color when tubers were pretreated with 1-MCP (Prange *et al.*, 2005). The authors hypothesized that 1-MCP could bind to the ethylene receptors and that the continuously applied ethylene regulated sprout growth by binding to newly formed ethylene receptors in the sprout eyes, where mitotic activity is highest. The authors further suggested that at sites of high mitotic activity in addition to the production of new ethylene receptors, greater 1-MCP metabolism may occur, since Huber *et al.* (2010) suggested that 1-MCP may be metabolized *in planta*. It should be noted that there is little supporting evidence for 1-MCP metabolism.

### ***Other effects of 1-MCP on potato***

There have been no reports of 1-MCP affecting sprouting of potato. There was no significant effect of 1-MCP on either applied a single dose or applied on multiple occasions on sprout length of any variety tested by Colgan *et al.* (2013).

In the absence of ethylene, 1-MCP had an effect on sugar levels, reducing glucose, fructose and sucrose in Cabaret, and glucose in Hermes for up to 4 months storage. There was no observable effect with other varieties including Maris Piper, Russet Burbank and Saturna (Colgan *et al.*, 2013)

### ***Storage temperature, ethylene and 1-MCP efficacy***

The effect of ethylene on fry colour and sugar levels appears greater at 6°C than 9°C (Colgan *et al.*, 2013). Foukaraki (2013) detected significant changes in reducing sugars on ethylene treatment in varieties stored at 6°C. 1-MCP may have greater impact on ethylene induced effects at the lower temperature.

## **5. Practical application of 1-MCP treatment for potato storage**

### ***Health and Safety and 1-MCP treatment***

As far as is known there are no significant concerns to commercial treatment. At standard temperature and pressure 1-MCP is a gas that decays, dissipates or is diluted in air such that there is no or negligible residue within 24 hours of application. Other than the standard care required in the handling of all chemicals, no additional care or protection is required if airborne concentrations are maintained at, or below, the specified treatment levels.

### ***Potato store filling and timing of application***

Commercial stores can be very large, of the order of 1000 tonnes and 1500m<sup>3</sup> volume. Store filling may typically take one to many weeks depending on harvest conditions *etc.* and this will impact on the potential to treat as early as possible after harvest.

### ***Size and integrity of Store***

1-MCP application into small contained areas or tightly sealed controlled atmosphere stores is relatively straightforward. Difficulties can be envisaged in achieving and maintaining an adequate 1-MCP concentration for sufficient duration in some of the potato stores currently in use. Potato Council-funded study R439 (2011) found potato stores to have gaps (equivalent leakage areas) within the structure equivalent of 0.33 - 5.5 m<sup>2</sup> with consequent difficulties in maintaining a sealed environment for the duration of treatment. It is possible that the effective area to be treated, and leakage, could be reduced by temporary sheeting.

### ***Cost/benefit of 1-MCP use***

Currently, 1-MCP is not registered for use on potato in GB or Europe. A business case decision will be needed to understand how costs for the work required for approval submission could be recouped by subsequent sales of the product. At the time of writing, 1-MCP, both as an active and its application formulations, are under patent. These lapse over the following few years but it seems unlikely that there will be a significant change in the cost of treatment in the foreseeable future.

The cost-benefit is difficult to estimate in the absence of likely commercial 1-MCP application costs. Treatment of apple stores costs in the region of £12/tonne. Depending on business case decisions the cost structure for potatoes could be lower. However, typically potato stores have a larger headspace than found in apple stores. Headspace must be taken into account as it is the entirety of store air volume that must be treated.

Studies to date have found an effect of 1-MCP in mitigation of ethylene induced adverse fry colour and sugar level changes. However, the effects of ethylene treatment are variety dependent and there is also an effect of temperature, at 9°C the effects on fry colour appear reduced compared to lower temperatures. For some processing varieties the effect of ethylene on fry colour is relatively small or absent (Colgan *et al.*, 2013) and in most varieties effects decline somewhat with storage duration. Ethylene treatment affects crisp fry colour more significantly than French fry colour. The effect of 1-MCP treatment is also variety dependent and this limits the benefits of 1-MCP to a sub-set of processing crops. For selected varieties treatment could be balanced against the benefit of reduced fry colour.

### ***Future developments or approaches***

AgroFresh offer a number of products that extend the range of use of 1-MCP. Harvista™ technology is a pre-harvest sprayable formulation that can be used to manage fruit ripening in the orchard. An AgroFresh-Syngenta product Invinsa™ is another sprayable formulation targeting protection of field crops during periods of stress e.g. drought. These products are targeted at foliage and the effects on root crops are not known.

It appears that the earlier the 1-MCP treatment the more beneficial results may be. A number of factors may constrain very early or effective application in commercial stores. The availability of a sprayable formulation has potential on-harvester use with benefits of early application, improved efficacy and possibly stress reduction

Other potential effects of 1-MCP treatment include both negative and positive effects on the development of physiological and pathological disorders (Watkins 2006). There is as yet no information on the possible role of 1-MCP for these disorders for potato. It is possible that effects on reduction of stress cf. Invinsa™ would alter the cost/benefit balance.

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