

Bud Fall Induction in Clove (*Syzygium Aromaticum*)

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ABSTRACT

*The abstract is to be in fully-justified italicized text, at the top of column as it is here, Cloves - the dry unopened flower buds of *Syzygium aromaticum* - represent the main source of income for many farmers in Indonesia, Tanzania, India, and Sri Lanka. The main issues associated with this crop are the pronounced biennial, triennial and quadriennial bearing, which lead to irregular pattern of production, and the very high costs of production. The objective of this experimental work was to induce the bud fall by applying hormone-like chemicals, precursors of the abscisic acid, which is the hormone responsible for the separation of leaves, flowers, fruits and other parts of the shoots at maturity or senescence. From a business perspective, the goal was to decrease the costs of harvesting, which represent more than 40% of the total production costs. In fact, the chemical induction of the buds would have allowed for the synchronization of bud harvest and eliminated the need for hand-picking buds from tall trees. Two different chemical compounds were tested (ACC and Ethephon) at varying concentrations. The experiment did not produce the expected results, as the flower buds fall was not induced. Several technical, ecological and physiological reasons can be taken into account to explain such results.*

Keywords: Chemical abscission, *eugenia caryophyllata*, harvesting, sustainable agriculture, thinning.

INTRODUCTION

Syzygium aromaticum (L.) Merr. et Perry is an evergreen tree belonging to the *Myrtaceae* family. It is cultivated for the production of cloves, dried unopened flower buds which are used as spices in gastronomy and as the essential part for kretek cigarettes (Polzinet *al.*, 2007). Clove tree is cultivated in the islands of Tanzania (Pemba and Zanzibar), Madagascar, Indonesia, Comoros Islands, and Sri Lanka, but the major producer is Indonesia, with 50.000 – 60.000t per annum (Leela and Sapna, 2008). Tobacco industry in Indonesia employs about 11 million workers and is the second largest employer after the government (Nichteret *al.*, 2009); cigarettes represent an important source of national revenue for Indonesian Government, whose tobacco taxes were approximately 38 trillion rupiah (\$4.2 billion US dollars) in 2006 (Graham, 2006). The tobacco market in Indonesia is unique because over 90% of all smokers smoke *kretek* cigarettes, and only a 10% smoke “white” cigarettes. Modern-day *kreteks* consists of Indonesian-grown tobacco (60-85% by wt.), chopped clove buds (15-40% by wt.) and a brand-specific flavoring spices mix (Hanusz, 2000).

S. aromaticum is a tropical plant that requires warm and humid climate, with an annual rainfall of 2500-3000 mm. It has no altitude requirements, as it can grow from sea level to 1000 meters. Clove is cultivated as 7 x 7 m) or consociated crop with coconut palm, areca nut palm, pepper, coffee and banana. It is a perennial plant, reaching 30 m in height, starting to bear flowers after 5 years and reaching the full production after 15-20 years (Martin *et al.*, 1987). Harvesting is manual: flower buds are hand picked using step ladders without damaging the branches, as this would adversely affect the succeeding growth. The buds are hand separated from the cluster after segregation from the stalks, and evenly spread to

facilitate sun drying, on mats or cement floors. At night, buds should be stored undercover, to avoid moisture re-absorbing. The period of drying depends on the climatic conditions and is typically accomplished four or five days under direct sun while it takes about four hours in artificial drying conditions (desiccator).

The total production of clove witnessed a decrease in the last 10 years due to some agronomic, economic and social issues which are harshly affecting the farmers, inducing many of them to shift to other less problematic crops (cocoa, banana, palm, cassava). From a merely agronomic point of view, the main issues regard, in particular, the irregular temporal pattern of production, with biennial, as well as triennial and quadriennial bearing (De Waard, 1974). This problem is particularly important in Indonesia, with remarkable triennial and quadriennial fluctuation depending on the total year rainfall, the exposure to low and high temperatures, the length of the dry season and other ecological and physiological parameters.

However, the main problem is associated with the harvest of the intact flower buttons which requires a very skilled and time consuming manual picking followed by their segregation from the stalks and from the cluster. Even though the juvenile phase of clove trees lasts for about 5 years (a relatively short time in fruit trees), the full production is reached only when the plants are very high (20 m) and very vigorous, leading to a difficult, slow, and dangerous harvesting processes. Moreover, the determination of the precise picking time is a crucial to optimize the yield, the concentration of essential oils in the dried product [according to several recent literature references (Ayoolaet al., 2008, Alma et al., 2007, Polzinet al., 2007, Jirovetzet al., 2006, Raina et al., 2001), 18 compounds represent more than 99% of the essential oil from clove. The major components are as follows: eugenol, 87%, eugenyl acetate, 8%, β -caryophyllene, 3,5%] and the following year flowering time and bearing of the tree (Martin et al., 1988); in fact, all the cloves on a tree seldom mature at the same time and, to harvest the entire crop, trees must be picked several times during the harvesting season. From an economic point of view, harvesting represents, in Indonesia, up to 40% of the total production costs, depending on labor availability and location.

For these reasons, the main goal of the experiment was to evaluate the application of some hormone-like chemicals as a tool to induce in a controlled way the flower buds fall.

The chemical thinning of flower buds is difficult to achieve, and little bibliographic records are available, also because only few plant species are cultivated for the production of the unopened flower buttons (buds). One of them, for instance, is *Capparis spp.*, which is cultivated for the production of capers, the brined unopened flower buds. Capers are manually harvested as well, but efforts to obtain a more mechanized harvesting of *Capparis spp.* have all failed (Tuttolomondoet al., 2006).

Several publications describe the application of growth regulators to fruit trees of the *Rosaceae* family and to grape, whose flower buds are sometimes too many to provide a sufficient development of each single fruit. In this cases, chemical thinning is usually obtained by means of molecules that induce the drop of flower buds of fruitlet, such as hydrogen cyanamide, dinitro-orthocresol, ammonium thiosulphate, naphthaleneacetic acid, BA, Carbaryl, and 2-chloroethylphosphonic acid. Unfortunately, all these treatments are aimed at reducing the *liveability* of the fruit buds, not their abscission. Sometimes they fall, but only after a deep physiological modification which, in most cases, totally compromises the characteristics of the flower bud itself.

The most interesting results on buds fall were obtained when using the endogenous ethylene naturally produced by the plant, or by applying natural or chemical precursors of the ethylene. The application of this plant hormone is a known effective tool since the 70's, and gave good

results on flowers and floral buds abscission: Veliath and Ferguson (1973) applied ethephon (which is converted in ethylene by the plant metabolism) at 1000 ppm and caused the first appreciable bud drop in tomato plants. More recently, YanChang *et al.* (2003) obtained comparable positive results on the abscission of the distal areal of the pedicel of the same species by applying very small quantities of ethylene to the plants. Another early and interesting study was done on *Phaseolus vulgaris* L. (Webster *et al.*, 1975): the application of 250 ppm ethephon promoted bud and flower abscission, while leaf abscission was unaffected as well as the total number of fruit and seed of the following season.

Nakamura and Wakasugi (1978) studied the effects of the timing of ethephon application on persimmon tree: when sprayed at the flower bud stage, flowers abscised at the juncture between calyx and peduncle, whereas when sprayed at the young fruit stage, they abscised at the juncture between calyx and fruit.

Ethylene has a great effect on the abscission of ornamental cut-flowers, in particular when administered during transportation: in 1987, Woltering investigated the sensitivity of more than 50 ornamental plants species to the exposure of exogenous ethylene, finding that 1-15 $\mu\text{l/l}$ ethylene caused the abscission of flower buds and flowers (at various stages of development) after 24 hours.

In 1991, Ethrel (ethephon) was applied at various concentration to sweet peas plants (Ohwaka *et al.*): the 100 ppm Ethrel treatment resulted in the abscission of a large number of developing flower buds, which showed a rapid rate of endogenous ethylene production immediately after spraying.

The effects of ethylene exposure of members of the *Myrtaceae* family was recently investigated (Macnish *et al.*, 2005): the separation at a morphological and anatomically distinct abscission zone between the pedicel and floral tube of an opened flower was already obtained when using only 1 $\mu\text{l/l}$ ethylene for 6 h induced, whereas 10 $\mu\text{l/l}$ for 24 h were necessary to completely abscise the flower buds enclosed in shiny bracteoles. It is even true that there is no bibliographic evidence on the same effects on clovetree or on the morphological and physiological characteristics of the fallen buds. It would be thus interesting to investigate the abscission power of ethylene and ethephon on clove tree, and the possibility to spray those chemicals to support the flower buds fall.

Though the application of ethephon-based chemicals was deeply investigated and easily obtained via canopy spraying, treatments with ethylene are much more difficult to obtain, as they require a controlled environment and/or time-prolonged applications (24 to 48 hours). For this reason, we envisioned the possibility of using different ethylene-releasing substances or ethylene-precursors such as 1-aminocyclopropane-1-carboxylic acid (ACC). Direct applications of ACC solutions to various parts of the flower of some species (*Citrus limon*, *Pelargonium* spp.) led to an increased ethylene production in the specific flower part (Hiloti *et al.*, 2000).

MATERIAL AND METHODS

The experiment was conducted on a private clove plantation (PT. PerkebunanCengkeh Zanzibar) in Curug Semarang (7°01'42''S 110°42'28''E), Central Java, Indonesia.

Bud fall induction was studied on 56 adult clove trees (*Syzygium aromaticum* (L.) Merr. et Perry). Individuals were chosen throughout the selected plantation during the first survey in the field (January 2009); the trees were all in very good shape, free from any visible disease of biotic or non-biotic origin. The trees were similar in height (about 15 m), LAI index and plantation year (1974). As the chemicals application timing is crucial for the evaluation of the

buds fall, a second survey was conducted in April 2009 and allowed to check proper and similar maturation stage of the buds of the selected trees (Figure 1).



Figure 1. Flower buds of clove tree at proper stage of maturation

Each treatment was applied on 8 individuals following a randomized blocks design of experiment. Ethephon (CAS 16672-87-0) and ACC (CAS 22059-21-8) were tested for their effects on cloves (buds) abscission. Following a literature survey, the following concentrations were tested: Ethephon 100, 250, 500 and 1000 ppm; ACC 10^{-6} , 10^{-5} and 10^{-4} M (table 1).

Table 1. Treatments applied in the study

Chemical	No. of samples	Concentration	Application	Water per plant (l)
Ethephon	8	100 ppm	1 mg/l	20
Ethephon	8	250 ppm	2,5 mg/l	20
Ethephon	8	500 ppm	5 mg/l	20
Ethephon	8	1000 ppm	10 mg/l	20

RESULTS AND DISCUSSION

After the applications, plants were strictly observed for five days in order to evaluate: 1. the date (or hours after chemicals application) of the first bud fall; 2. the percentage of fallen buds after three, six, twelve and twenty-four hours after the treatment; 3. the total percentage

of fallen buds; 4. the initial and the total percentage of fallen leaves; 5. the presence of necrosis (black or brown spots or burns) or chlorosis on fallen/not fallen leaves, their initial and total percentage, their evolution throughout the five days of observation.

After a negligible initial bud fall, probably due to the mechanic impact of the sprayed liquid, we did not appreciate any buds fall during the day one nor during the following days.

The chemical analysis on the fallen buds, in order to evaluate the presence of potentially toxic chemical residuals, and the presence of essential oils in sufficient concentrations wasn't done on the fallen buds, nor on the buds still on the trees due to the lack of expected bud fall.

The in-depth bibliographic research performed on the agronomic techniques applied on clove tree has revealed a very lacunose global knowledge of this tropical crop. Few studies are available, namely concerning the essential oils of the flower buds and the genetic breeding on some cultivars. Very few experiments have been performed in the field in order to address the problems related to bearing fluctuation and labor effort, as well as economic issues related to the manual harvesting of the flower buds. The clove harvestable part is the unopened flower buttons, and this peculiarity is shared only with *Capparis* spp. (in southern Italy and in some other arid zones as Turkey)

The manual harvest of any plant part (leaves, shoots but, in particular, flowers and fruits) represent an obstacle to the extensive cultivation of lots of species (*Pyruscommunis*, *Prunusmalus*, *P. persica*, *P. armeniaca*, *P. avium*, *Rubus* spp., *Oleaeuropea* etc.), and in all cases the labor effort increases the production costs and decreases the income per hectare. Published studies demonstrate that the scientific community has worked hard to address this aspect, usually suggesting agronomic techniques or the application of chemicals to obtain the natural fall of fruits in cherry (Kollar and Scortichini, 1986), and apple (Schumacher and Stadler, 1993), two fruit trees where the harvest is mechanically assisted.

The negative results reported here can be explained by one or more of the following reasons: 1. the concentration of the sprayed solutions was not optimized; 2. the sprayed precursors of abscisic acid did not reach the right tissues and the synthesis of a sufficient concentration of abscisic acid in the abscission zones was not triggered; 3. the fall of the buds should have been followed by a mechanical shaking of the trunk; 4. the wind drift prevented the solutions from reaching the target tissues. These are only some of the ecological, physiological and practical speculations that could explain the failure of the experiment, and which thus deserve further investigations. Some other useful tests to perform in the future (next flowering season or in the next two or three years) should be the following: (1) Increase of the concentrations of ACC and Ethephon in solution; (2) Selection of younger or shorter (dwarf) samples in order to obtain a very good application of the chemicals; (3) Selection of some other chemicals to test that could be effective in the flower buds fall.

From a technical point of view, in order to facilitate the experiment and to test as many concentrations and chemicals as possible, it should also be feasible to select only some parts of the tree, to address the applications to the lowest branches. These samples would require smaller volumes of solutions, which would be applicable without using high-pressure spray pumps but small hand spray tools. The possible wind drift would also be prevented. In case one or more applications induced the desired buds fall, the selected chemical at the effective concentration would then be applied (the same year or the following year) at a tree scale in order to evaluate the feasibility of the treatment.

CONCLUSIONS

On order to solve the problem of manual harvesting of clove flower buds, and to lower the cost production of the spice, this research was focused on obtaining the chemical abscission of flower buds applying two hormone precursors of abscisic acid (ACC and Ethephon). As no bibliographic reports were found on chemical thinning on clove trees, the concentrations and application methods of chemicals were chosen basing on the studies developed on other species, in particular in order to obtain the chemical thinning of fruit lets. As no results were obtained, further studies are necessary to achieve the economic results.

These new studies could be focused on:

- testing new hormone precursors or other chemicals on selected parts of the same samples, or different (higher) concentrations of the same hormones already used
- testing several chemical thinning agents on another species, a small tree or a shrub, whose harvest is the flower button, reducing the experimental economic and labour costs
- applying growth regulators and bending the shoots of young clove tree samples in order to obtain shorter stems and internodes, juvenility reduction, higher flower bearings and to limit the biennial, triennial or quadrennial production fluctuation.

REFERENCES

- [1] Feldman, R. S. (1996). *Understanding Psychology*. Newyork: McGraw-Hill,Inc.
- [2] Alma et al., (2007). Research on essential oil content and chemical composition of Turkish clove (*Syzygium aromaticum* L.). *Bio Resources*, 2(2): 265-269.
- [3] Ayoola et al., (2008). Chemical analysis and antimicrobial activity of the essential oil of *Syzygium aromaticum* (clove). *African Journal of Microbiology Research*, 7(2): 162-166.
- [4] Biswas, B. (1994). Effect of growth substances on growth, flowering and fruiting of papaya. *Annals of Agricultural Research*, 15(3): 301-305.
- [5] Dalal et al., (2005). Effect of chemical on flowering and fruit yeld of mango cv. Pairy. *International Journal of Agricultural Sciences*, 1(1): 24-25.
- [6] De Waard, P.W.F. (1974). The development of clove loads and causes of irregular bearing of cloves [*Eugenia caryophyllus* (Sprengel) Bullok et Harrison]. *Journal of Plantation Crops*, 2(2): 23-31.
- [7] Dutta et al., (2008). Effect of plant bio-regulators on fruit quality and mineral composition of ripe mango cv. Himsagar. *Indian Agriculturist*, 52(3/4): 107-111.
- [8] Hanusz, M. (2000). *Kretek, The Culture and Heritage of Indonesia's Clove Cigarettes*. Equinox Publishing, Tortola, British Virgin Islands.
- [9] Hilioti et al., (2000). Regulation of pollination-induced ethylene and its role in petal abscission of *Pelargonium * hortorum*. *Physiologia Plantarum*, 109: 322-332.
- [10] Jirovets et al., (2006). Chemical composition and antioxidant properties of clove leaf essential oil. *Journal of Agricultural Food Chemistry*, 54: 6303-6307.
- [11] Kollar, G., & Scortichini, M. (1986). Effetti di trattamenti chimici facilitanti l'abscissione del frutto della cultivar di ciliegio dolce "Germesdorf". *Rivista di Ortoflorofrutticoltura italiana*, 70: 85-95.

- [12] Leela, N. K., & Sapna, V. P. (2008). *Clove*. In: Parthasarathy, V.A., Chempakam, B., Zachariah, T.J. (Eds). *Chemistry of spices*. CAB International, Cambridge, USA, pp. 146-164
- [13] Macnish et al., (2005). Anatomy of ethylene-induced floral-organ abscission in *Chamelaucium uncinatum* (Myrtaceae). *Australian Journal of Botany*, 53: 119-131.
- [14] Martin et al., (1988). Causes of irregular clove production in the islands of Zanzibar and Pemba. *Experimental Agriculture*, 24: 105-114.
- [15] Martin et al., (1987). Clove tree yields in the islands of Zanzibar and Pemba. *Experimental Agriculture*, 23(3): 293-303.
- [16] Menon, P. S. (2000). IISR scientists develop new clove varieties. *Journal of Herbs, Spices & Medicinal Plants*, 7(1): 103-105.
- [17] Mukhopadhyay, A. K. (1976). A note on the effect of growth retardants and L-methionine on flowering of mango (*Mangifera indica* L.). *Haryana Journal of Horticultural Sciences*, 5(3/4): 169-171.
- [18] Nakamura, M., & Wakasugi S. (1978). Chemical thinning of Japanese persimmon trees using Ethrel sprays. I. The influence of Ethrel on the formation of the abscission layer of the flower or fruit and its development. *Journal of the Japanese Society for Horticultural Science*, 47(3): 308-316.
- [19] Nichter et al., (2009). Reading culture from tobacco advertisements in Indonesia. *Tobacco control*, 18: 98-107.
- [20] Polzin et al., (2007). Determination of eugenol, anethole, and coumarin in the mainstream cigarette smoke of Indonesian clove cigarettes. *Food and Chemical Toxicology*, 45(10): 1948-1953.
- [21] Raina et al., (2001). Essential oil composition of *Syzygium aromaticum* leaf from Little Andaman, India. *Flavour and Fragrance Journal*, 16(5), 334-336.
- [22] Sarkar et al., (1998). Regulation of tree vigour in mango. *Indian Journal of Horticulture*, 55(1): 37-41.
- [23] Schumacher R. & Stadler W. (1993). Fruit set regulation and quality. *Acta Horticulturae*, 326: 49-57.
- [24] Singh S. P., & Borase S. S. (2003). Extending shelf-life of tropical fruits through ripening retardants. *Advances in Horticulture and Forestry*, 9: 79-100.
- [25] Tuttolomondo et al., (2006). Origano, rosmarino, timo, mirto e capperò: caratterizzazione, propagazione e tecniche colturali. *Informatore Agrario Supplemento*, 62(50): 21-25.
- [26] Veliath J.A. & Ferguson A.C. (1973). A comparison of ethephon, DCIB, SADH, and DPA for abscission of fruits, flowers, and floral buds in determinate tomatoes. *Journal of the American Society for Horticultural Science*, 98(1): 124-126.
- [27] Webster et al., (1975). Effects of ethephon on abscission of vegetative and reproductive structures of *Phaseolus vulgaris* L. *Hort Science*, 10(2): 154-156.
- [28] Woltering E. J. (1987). Effects of ethylene on ornamental pot plants: a classification. *Scientia Horticulturae*, 31(3/4): 283-294.
- [29] YanChang et al., (2003). Effect of ethylene on abscission of tomato pedicel in vitro. *Acta Horticulturae Sinica*, 30(5): 554-558.