**UNIVERSITI PUTRA MALAYSIA**

# EFFECTS OF CHITOSAN IN EXTENDING THE VASE LIFE OF CUT CHRYSANTHEMUM (DENDRANTHEMA MORIFOLIUM RAMAT) FLOWERS

**EDDY AZIAN.**

# EFFECTS OF CHITOSAN IN EXTENDING THE VASE LIFE OF CUT CHRYSANTHEMUM (*DENDRANTHEMA MORIFOLIUM* RAMAT) FLOWERS

By

EDDY AZIAN

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfilment of the Requirements for the Degree of Master of Science

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*...... Ya Allah, ya Tuhan, aku berlindung kepadaMu dari ilmu yang tidak berguna, dari hati yang tidak tunduk, dari nafsu yang tidak puas, dan dari permohonan yang tidak diterima. (H.R. Muslim)*

**SPECIAL DEDICATIONS**

This Thesis is dedicated to

my beloved parents, Zahari Nur and Hilalliah

my beloved parents in law, Supriatin and E. Muchtar

my beloved wife, Neni Mulyani, whose patience, supports and companionship has facilitated my work,

my daughter, Miza Hilmiya

Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Master of Science

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**Chairman:** **Associate Professor Mohamad Zaki Ab. Rahman, PhD**

**Faculty:** **Science**

The effect of the isolated bacteria strains on the vase life of cut flowers of Chrysanthemum (*Dendranthema morifolium* Ramat) was investigated. The bacteria strains were isolated from the cut stem end and vase water of cut chrysanthemum flowers. The longevity of vase life and total water uptake of cut chrysanthemum was reduced by placing the stems in a suspension of *Pantoea agglomerans* and *Burkholderia multivorans* at $10^8$ CFU $mL^{-1}$.

The study was also conducted to look at the effects of chitosan in extending the vase life of cut chrysanthemum. Chitosan was applied as pulse treatment. Cut flowers were treated with 1) chitosan at 0, 25, 50, 75 and 100 mg $L^{-1}$, 2) chitosan at concentration 1) supplemented with 2% sucrose, 3) chitosan at concentration 1) supplemented with 1.5 mM $Ca(NO_3)_2$ and 4) chitosan at concentration 1) supplemented with 1.5 mM $Ca(NO_3)_2$ and 2% sucrose. The experiment was

carried out in the Postharvest Laboratory with the environmental conditions, 25 ± 2°C, 70% RH and 15 μmol $m^{-2}$ $s^{-1}$ light intensity from cool-white fluorescent lamps for 12 hours. Compared to those held in control, all treatments extended the vase life. Treatment 75 mg $L^{-1}$ chitosan supplemented with 1.5 mM $Ca(NO_3)_2$ was the most effective in extending the vase life.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

**KAJIAN TERHADAP PENGGUNAAN KITOSAN UNTUK MELANJUTKAN HAYAT JAMBANGAN BUNGA KERATAN KEKWA (*DENDRANTHEMA MORIFOLIUM* RAMAT)**

Oleh

**EDDY AZIAN**

**Mac 2006**

**Pengerusi:** **Profesor Madya Mohamad Zaki Ab. Rahman, PhD**

**Fakulti:** **Sains**

Kesan strain bakteria yang diasingkan terhadap hayat jambangan bunga keratan kekwa (*Dendranthema morifolium* Ramat) telah dikaji. Strain bakteria diasingkan daripada pangkal batang dan cecair pasu bunga keratan kekwa. Tempoh hayat jambangan dan jumlah serapan air bunga keratan kekwa berkurangan apabila batang bunga kekwa diletakkan dalam suspensi *Pantoea agglomerans* dan *Burkholderia multivorans* pada populasi $10^8$ CFU $mL^{-1}$.

Satu kajian lagi juga dilakukan untuk melihat kesan kitosan dalam memanjangkan hayat jambangan bunga keratan kekwa. Kitosan diperlakukan sebagai rawatan *pulsing*. Bunga keratan kekwa dirawat dengan 1) kitosan pada kepekatan 0, 25, 50, 75 dan 100 mg $L^{-1}$, 2) kitosan pada kepekatan 1) dengan tambahan 2% sukrosa, 3) kitosan pada kepekatan 1) dengan tambahan 1.5 mM $Ca(NO_3)_2$ dan 4) kitosan pada kepekatan 1) dengan tambahn 1.5 mM $Ca(NO_3)_2$ dan 2% sukrosa. Kajian dilakukan dalam makmal pascatuai dengan suhu persekitaran 25 ± 2°C,

70% RH and 15 µmol $m^{-2}$ $s^{-1}$ cahaya lampu putih selama 12 jam sehari. Jika dibandingkan dengan kawalan, semua rawatan memanjangkan hayat jambangan. Dengan rawatan 75 mg $L^{-1}$ kitosan bersama 1.5 mM $Ca(NO_3)_2$ memberikan pemanjangan hayat jambangan yang terbaik.

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## TABLE OF CONTENTS

## LIST OF TABLES

## LIST OF FIGURES

# LIST OF ABBREVIATIONS

| | |
|---|---|
| AIB | aminoisobutyric acid |
| ANOVA | analysis of variance |
| atm | atmosphere |
| BCDMH | 1-bromo-3-chloro-5,5-dimethylhydantoin |
| °C | degree centigrade (celcius) |
| CFU | colony forming unit |
| cm | centimetre |
| CRD | completely randomized design |
| DICA | dichloroisocyanuric acid |
| DMRT | Duncan's multiple range test |
| e.g. | for example |
| *et al.* | et alteri |
| g | gram |
| GlcN | *N*-acetyl-β-D-glucosamine |
| GN | gram negative |
| ha | hectare |
| HQC | hydroxyquinoline citrate |
| klx | kilolux |
| KOH | potassium hydoxide |
| L | litre |
| μ | micro ($10^{-6}$) |
| M | molar |

| | |
|---|---|
| m | milli ($10^{-3}$) |
| MIC | minimum inhibitory concentration |
| MT | metric ton |
| MW | molecular weights |
| ppm | part per million |
| RH | relative humidity |
| s | second |
| SEM | scanning electron microscopy |
| ST | simulated transport |
| STS | silver thiosulphate |
| v/v | volume / volume |
| w/v | weight / volume |
| w/w | weight / weight |

# CHAPTER I

# INTRODUCTION

The vase life longevity is one of the most important quality factors for cut flowers (Pun *et al.*, 1999; Ichimura *et al.*, 2002). Most of cut flowers show leaf wilting shortly after being placed in the vase solution. Cut flowers are kept at room temperature in houses for decoration and end up dry mostly through water loss (Anjum *et al.*, 2001). This water stress develops soon after placing the flowers in water and this often cause shortening of vase life. Unrestricted water uptake through the xylem conduits is of paramount importance in providing an adequate supply of water to transpire cut stems. According to Havely and Mayak (1981) and van Doorn (1997), the main reason for water stress during the vase period is resistance to water flow which develops in the stems.

According to van Doorn *et al.* (2002) the water uptake during vase life of flowers can be shortened, due to development of an occlusion in the xylem conduits, leading to premature wilting of the leaves and flowers. There are several causes of occlusion, such as microbial contamination in the water and microbial growth in the stems, and a physiological response for cutting.

The presence of bacteria in vase water greatly influences the longevity of cut flowers. This was observed in cut carnation (van Doorn *et al.*, 1995), roses (Lineberger and Steponkus, 1976; de Witte and van Doorn, 1988; van Doorn and

Perik, 1990; Florack *et al.*, 1996), and *Adiantum raddianum* frond (van Doorn *et al.*, 1991). Besides that, the presence of microbial in vase water can also cause release of toxic metabolites and/or enzymes, and evolve damaging levels of ethylene (Gilman and Steponkus, 1972; van Doorn and Perik, 1990; Jones and Hill, 1993).

The use of floral preservative is the most economical and practicle method of extending the postharvest life of cut flowers. Several preservative such as silver thiosulphate (STS), silver nitrate ($AgNO_3$), and aluminum sulphate have physiological effects on vase life and have been reported to promote flowers fresh weight increase, reduced microbial numbers, and inhibited the effects of ethylene. However, chemical control program causes environmental problems, such as, an increasing number of fungicides-resistant strains of postharvest pathogens, and a number of commonly used fungicides are under review due to health risk concerns (El Ghaouth *et al.*, 1991; Florack *et al.*, 1996). Thus, exploring ways to replace these chemical compounds devoid of any heavy metal (Onozaki *et al.*, 1998) by natural products with antibacterial properties or to intensify the natural defences of the tissue to control decay and prolong storage life is receiving more and more attention (Florack *et al.*, 1996; Zhang and Quantick, 1998).

Chitosan is a linear polymer composed of β-1,4-linked glucosamine (GlcN) with various degrees of N-acetylated GlcN residues, is a deacetylated derivative of chitin extracted from an abundant source of shellfish exoskeletons. The amino polysaccharide and its derivatives have many diverse applications in agriculture, medicine and cosmetics (Kittur *et al.*, 2003).

The use of a natural substance such as chitosan has been considered as a valid alternative to synthetic fungicides (Romanazzi *et al.*, 2003) because the polymer which is naturally occur should not cause health and pollution problems. The unique positively charged chitosan in acidic solution has potential uses in agriculture such as a carrier for the controlled release of fungicides and fertilizers. According to Jung *et al.* (1999), antifungal or antimicrobial activities of chitosan are believed to originate from the polycationic nature of chitosan that can be bind with anionic sites in proteins, thus resulting in selective antimicrobial activities towards fungi or bacteria.

Several recent studies have shown that chitosan has antibacterial and antifungal characteristics. Plant seeds soaked in aqueous solution of chitosan can prevent microbial infections and increased plant production. Ohta *et al.* (1999) has reported that chitosan promote growth of *Eustoma grandiflorum* seedlings and flowering.

Chitosan is used as spray or coating of fresh fruits and vegetables. Coating fruit with chitosan prolongs the storage life and better control of decay in tomatoes, and strawberry fruits (El Ghaouth *et al.*, 1992a and b), litchi, peach, Japanese pear and kiwifruit (Zhang and Quantick, 1997; Du *et al.*, 1997). Makino and Hirata (1997) reported that the biodegradable laminate chitosan-cellulose was able to use as packaging materials for storage of fresh vegetables (head lettuce, cut broccoli, whole broccoli, tomatoes, sweet corn and blueberries). Recently, Reddy *et al.* (2000) found that spray application of chitosan affected quality of strawberry fruits and having potential as an anti-transpirant of pepper (Bittelli *et al.*, 2001).

The use of edible films and coating to extend shelf life and improve the quality of fresh, frozen and food industry has been examined during the past few years (Kester and Fennema, 1986). The film-forming qualities of chitosan have been found to be dependent on the homogeneity of the bulk material, the degree of acetylation and acid degradation during dissolution. A composite film derived from chitosan have been developed with qualities such as good wet strength, excellent oxygen barrier properties and biodegradability (Hosokawa *et al.*, 1990; Conca and Yang, 1993; Caner *et al.*, 1998).

Most of the studies previously carried out only indicated that chitosan have been applied in postharvest treatments and preharvest application to extend storage life of fruits and vegetables. However, information of the use as cut flower preservation are still lacking. Therefore, the objectives of this study were:

1. To evaluate the role of bacterial strains in the vase life of cut chrysanthemum.
2. To evaluate the cut chrysanthemum flower quality by using chitosan supplements.