



Research article

Utilization of environmentally friendly essential oils on enhancing the postharvest characteristics of *Chrysanthemum morifolium* Ramat cut flowersIman Mohamed El-Sayed^a, Rasha Ahmed El-Ziat^{b,*}^a Department of Ornamental Plants and Woody Trees, Agricultural and Biological Research Division, National Research Centre (NRC), Egypt^b Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, Giza, Egypt

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ABSTRACT

Chrysanthemum is one of the most consumed and most valuable cut flowers worldwide. In this study, the effectiveness of three concentrations of either thyme oil (300,400 and 500 mg/l) or clove oil (150,250 and 500 mg/l) as additives in holding the postharvest solutions of chrysanthemum "Arctic Queen White" cut flowers were investigated. The experiments were carried out as a completely randomized design in three replicates. Many postharvest characteristics have been evaluated, such as the vase life of cut flowers, diameters of head flowers and stem, dry matter of flowers, total vase water uptake, total loss of water, relative fresh weight. Additionally, the chlorophyll contents, total sugar, and bacterial counts were determined. The results showed that the longest vase life of cut chrysanthemum was 36.50, 33.40 days, and 35.88, 31.33 days by addition of either the thyme oil (500 mg/l) or clove oil (250 mg/l) in holding solution as compared with distilled water (18.09 and 17.22 days) in both seasons. The highest total vase water uptake and relative fresh weight were (225.00, 211.05 g/flower/day) and (79.89, 70.37 %) of cut chrysanthemum treated with 500 mg/l thyme oil in both seasons. Whereas the lowest total water loss in the two seasons was 155.11 and 156.60 g/flower/day was found with 400 mg/l thyme oil. The greatest chlorophyll a, b, carotenoids, and total sugar contents obtained from treated cut chrysanthemum with 500 mg/l thyme oil (6.89, 2.37, 5.99 mg/g, and 0.88 mg/gm D.W, respectively). Furthermore, the treatment of cut flowers with selected oils has significantly decreased the bacterial growth compared to the control. Whereas the minimum bacterial activities were <1 C.F.U/ml with cut chrysanthemum fortified with 500 mg/l thyme and clove oils. Moreover, the superlative treatments with thyme (500 mg/l) and clove (250 mg/l) showed a prime state of xylem vessels comparable with the control. thus, the usage (addition) of thyme and clove oils as a natural preservative in holding solutions instead of chemicals would be of great economic and environmental impact (Values).

1. Introduction

Cut flowers are invaluable horticultural products. Preserving perfect quality cut flowers and prolonging the shelf life of cut flowers are essential and practical for having optimal products in markets (Redman et al., 2002; Macnish et al., 2008). The vase life of cut flowers is mainly affected by two essential factors, first, the ethylene, which accelerates the senescence of many flowers and microorganisms that cause vascular blockage resulting in reducing the shelf life of cut flowers (Zencirkiran, 2005, 2010). In addition, cut flowers are considered a source of national income, especially in countries with a suitable environment for growth and flowering. Egypt is one of these countries with many production

elements including, temperature, soil, light, and humidity, to cultivate and produce flower crops with high quality and quantity (Amin, 2017).

Chrysanthemum (*Chrysanthemum morifolium* Ramat) is one of the leading cut flowers in the international market, Queen of the East belongs Asteraceae family (Boukhebt et al., 2020). It is native to Southeast Asia and China and is widespread in Europe, North Africa, and America. It is also one of the most global five cut flowers with the highest economic importance for decoration and ornamental in the floriculture industry, and trades on the world markets as potted plants, cut flowers, and a background in borders by planting erect and tall varieties. Moreover, chrysanthemum is favored by many consumers in the flower market due to the variety of shapes and sizes of flowers (Dole and Wilknis, 2005). Nowadays, it represents the second-class economy in the world, and roses

* Corresponding author.

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followed. *Chrysanthemum* cut flowers are commonly used in two styles, standard (one flower on the stem) and spray (multiple flowers on the stem) (Khaligi, 2010; Amin, 2017).

Essential oils (EOs) are organic, natural products extracted from aromatic and medicinal plants and exhibit different antioxidant, anticarcinogenic, antibacterial, and antifungal properties, which may be used as natural additives in several plants and crops (Teissedre and Waterhouse, 2000; Bayat et al., 2013). Essential oils can be used in the preservative solution to control bacterial and fungal pathogens (Hegazi and EL Kot, 2009; Solgi et al., 2009; El-Hanafy, 2007). The essential oils have significant antimicrobial activities against some fungi and bacterial properties due to having high levels of phenolic compounds such as eugenol, carvacrol, and thymol (Bounatirou et al., 2007; Sharififar et al., 2007).

Thyme (*Thymus vulgaris*) is a member of the family Lamiaceae. Its oil mainly contains thymol, borneol, and carvacrol (Jakiemiu et al., 2010). It has antioxidative, antibacterial, and antimycotic properties (Imelouane et al., 2009). Solgi et al. (2009) reported that thyme oils are used in the vase solution for prolonging the shelf life of gerbera (*Gerbera jameson* cv. "Dune") cut flowers. Clove (*Syzygium aromaticum*) belongs to the family Myrtaceae and has many medicinal properties such as antimicrobial and antiviral activities (Osman et al. (2020); Devkota and Adhikar-Devkota, 2020; Osman et al. (2020); Cui et al., 2018). Clove plant is the main source of phenolic molecules such as hydroxybenzoic acids, flavonoids, hydroxyphenylpropenes, and hydroxycinnamic acids (Batiha et al., 2020). Also, the EOs collected from flowers, buds, stems, and the leaves of the plant, showed different chemical composition, colour, and flavour (Shahbazi and Shavisi, 2015). The essential oils of clove have been shown antimicrobial, anti-inflammatory, antioxidant, analgesic, antiparasitic, anti-convulsant, and anticarcinogenic properties (Ramadan et al., 2013; Sanuja et al., 2014; Lekjing, 2016).

The main aim of this study is the impact of thyme and clove essential oils as an eco-friendly preservative to improve post-harvest characters, especially vase life of cut flowers, water relations, chemical composition, and to evaluate their antibacterial activity on chrysanthemum cut flowers.

2. Material and methods

The experiment was carried out in the laboratory of the Ornamental Horticulture Dep., Fac. of Agric., Cairo Univ., Giza, Egypt, during the two successive seasons from 28 November 2018 to 3 January 2019. The study was performed to examine the influence of using some natural essential oils as preservative solutions on the vase life of chrysanthemum cut flowers and to improve their quality and to extend the shelf-life period. The Scanning electron microscopy was carried out in the National Research Centre (NRC).

2.1. Plant material

The cut flowers in this study were chrysanthemum (*Chrysanthemum morifolium* Ramat) cv, "Arctic Queen White". Cut flowers were obtained from the commercial growing farm "Floramix Farm" flowers were moved to the laboratory on the same day. The flowers were pre-cooled by placing in the cold water for 30 min after the stems were re-cut underwater to a length of 60 cm in all cut spikes and removed the leaves on the lower third of the stem. Every cut spike was placed individually in the vase (600 ml) containing vase solution 500 ml under 21 ± 1 °C and relative humidity $65 \pm 5\%$.

2.2. Essential oils treatments

Essential oils obtained from the unity of squeezing and extracting natural oils, National Research Centre (NRC). This experiment included examining the influence of thyme oil at the levels of 300, 400 and 500

mg/l of, and clove oil at 150, 250, and 500 mg/l. The distilled water (control) and 0.4% of sucrose as carbon source used as controls and were pulsed for the same period with distilled water. Tween-20 "0.1% v/v" was used to dissolve the oils before adding them to the preservative solution.

2.3. Data recorded

2.3.1. Vase life (day)

The vase life was calculated by counting the days since applying the treatment (first day) until wilting the leaves and the flower (Nabigol et al., 2005).

2.3.2. Diameter of head flowers (cm)

The diameter of head flowers was calculated as a mean of three randomly selected flowers per plant and assessed as marketable.

2.3.3. Diameter of stem (mm)

At a high 30 cm of the stem, the diameter of the stem was determined by using a digital caliper; an average of three stems per plant were selected randomly and calculated.

2.3.4. Dry matter

The fresh weight of flower stem of each treatment was determined at the end of flower vase life, and then it was dried to a constant weight in an oven for 24 h at 72 °C.

2.3.5. Total vase solution uptake (g/flower/day)

During the vase life evaluation time, the weighting of the vases containing water solutions without cut flowers was recorded every two days. Total days of the vase solution uptake rate were calculated at the end of the vase life period of each treatment by the following formula:

$$\text{Water uptake [g/flower/day]} = [St^{-1} - St];$$

where, St is the weight of water solution (g) at t = day 2, 4, 6, etc, and St^{-1} is the weight of water solution (g) on the previous day (0, 2, 4, 6, etc) by Bazaz et al. (2015).

2.3.6. Total water loss (g/flower)

Water loss was calculated during the vase life evaluation period. The weighting of the vases containing cut flowers and vase solution were recorded every two days. Total days of the vase solution loss rate were calculated at the end of the vase evaluation period of each treatment as;

$$\text{Water loss (g/flower/day)} = (Ct^{-1} - Ct);$$

where, Ct is the combination of the weight of cut flowers and the vase solution (g) at t = days 2, 4, 6 and Ct^{-1} is the combination of the weights of cut flowers and vase solution (g) on 0, 2 and 4 days, respectively, by Bazaz et al. (2015).

2.3.7. Relative fresh weight (RFW %)

The cut flowers were initially weighed at the beginning of the experiment and weighed again on days 0, 2, 4, and at the vase life of the control flowers. Relative fresh weight (RFW) of cut flowers was measured as described by He et al. (2006):

$$\text{RFW \%} = (Wt / Wt^{-0}) \times 100;$$

where, Wt is weight of flowers (g) at t = days 0, 2, 4, etc., and Wt^{-0} is weight of the same flowers (g) at t = day 0.

2.3.8. Chlorophyll content (mg/g F.W)

Chlorophyll content was obtained in the seven days during the shelf-life period. Chlorophyll a, b and carotenoids were determined according to Saric (1967).

2.3.9. Total sugar (mg/gm. D.W.)

Total sugar was recorded in the seven days during the shelf-life period. Total sugars of air-dried leaves were determined according to DuBois et al. (1956).

2.3.10. Averages of bacterial counts (C.F.U/ml)

Means of the bacterial population was determined in the keeping solutions after the 7th day. Where 1 ml was taken from each sample (300,400, and 500 mg/l of thyme oil, and clove oil at concentrations 150,250, and 500 mg/l. the distilled water (control)), diluted by using sterilized distilled water from the first dilution to the sixth dilution. After that, 1 ml of each fourth, fifth, and sixth dilution were inoculated in petri dishes on media containing agar, peptone, and beef extract then it incubated for 48 h at 30 °C, and the colonies have been counted by Marousky (1969).

2.3.11. Scanning electron microscopy (SEM)

Microscopic examination was used to scan xylem occlusion by microbes at the base of the cut flower stem. At the end of the shelf -life, sections (0.5 cm) were taken from the control flowers and treated flowers with 500 mg/l of thyme and 250 mg/l of clove oils. The cut faces were sealed with glue (conductive carbon glue or two-component epoxy glue). The xylem pieces were mounted on a scanning electron microscope (SEM) stubs with a conductive adhesive tab. The fresh samples were examined after metal coating (Ensikat et al., 2010).

2.4. Statistical analysis

The data were analyzed using a randomized complete design with three replicates per treatment. Each replicate consisted of one spike carrying 4–6 head flowers. The treatments average was compared for significance by new multiple range tests at the 0.05% level of probability (Duncan, 1955) using one way (ANOVA) COSTATV-63.

3. Results and discussion

3.1. Vase life

The influence of different treatments on vase life (day) using thyme and clove oils was determined. Data in Figure (1) Indicated that the addition of both thyme oil and clove oil in keeping solution was significantly increasing the vase- life of cut flowers. The longest mean on vase -life (day) was achieved using the preserved solution containing 500 mg/l thyme oil, which recorded (36.50 and 33.40 days, respectively) on both first and second seasons, compared with the control (distilled water) and other treatments. Similarly, the vase of cut flowers was 35.88 and 31.33 days in the first and second seasons, respectively, when 250 mg/l clove oil was preserved. However, the vase life's shortest mean value was recorded with added clove oil at 150 mg/l (22.17 and 19.15 days, respectively) in both seasons. Overall, the results indicated that the essential oils of both thyme and clove in the preservative solution significantly extended the vase life of chrysanthemum cut flowers compared to the control (distilled water).

These findings were confirmed by Solgi et al. (2009), who mentioned that oil of thyme enhanced the vase-life of cut flowers of *Gerbera jamesonii* and decreased bacterial contamination in the stem end. Bazaz and Tehranifar (2011) also extended the shelf-life of alstroemeria cut flowers using thyme oil. Similarly, Manfredini et al. (2017) mentioned that they use several oils, such as eucalyptus, cinnamon, lemongrass, and peppermint (1%) and clove oil (0.1%) enhanced floral stems in post-harvest of rose's cut flowers. Furthermore, the vase life of cut carnation was prolonged with the addition of thymol 25 or 50 mg/L (Solgi, 2018).

These findings may explain as the thyme contains several phenolics such as thymol, which have antimicrobial activities resulting in preventing or diminishing the bacterial growth, decreased the blockage of xylem tissues (Jalili Marandi et al., 2011; Di, 2008). Also, it improved the water uptake; prevent water stress, and wilting of flowers, which leads to

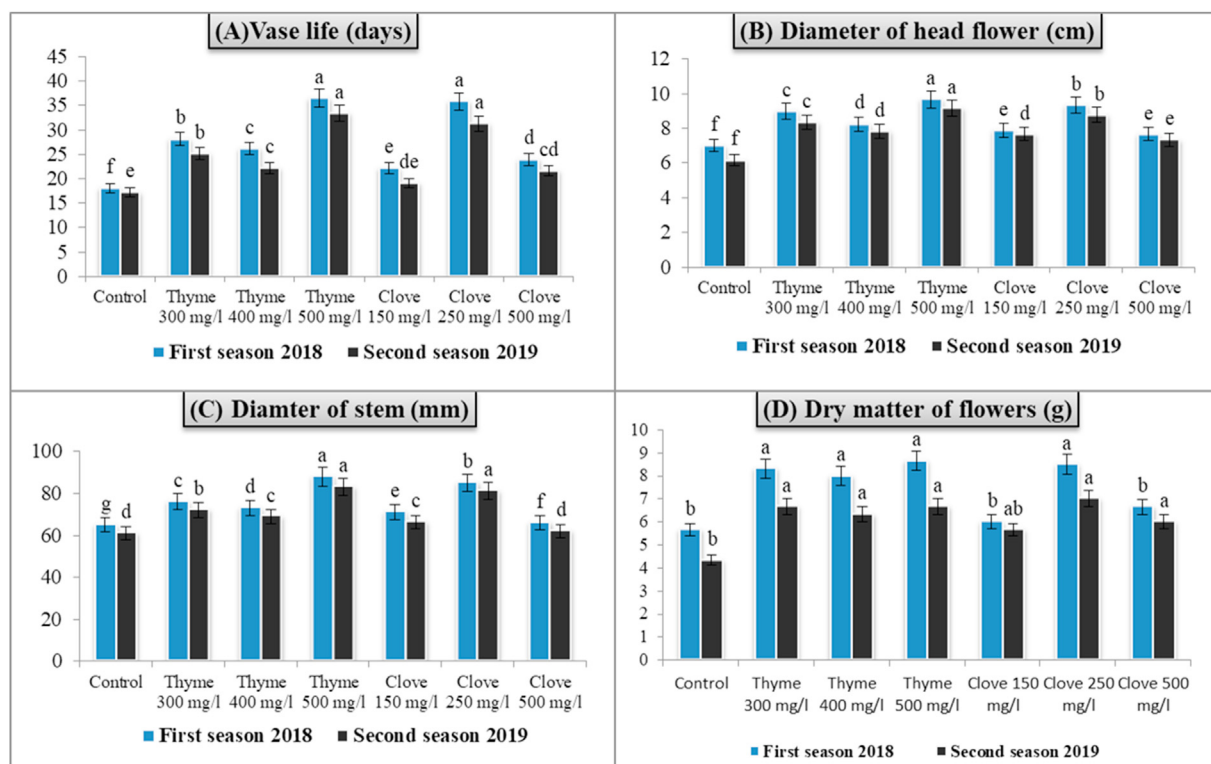


Figure 1. (A, B, C, and D) Effect of different concentrations of thyme and clove essential oils on flowering characteristics during 2018–2019 seasons. Columns data with different letter/s differ significantly ($p \leq 0.05$). letters (a–g) are comparison of individual treatment means.

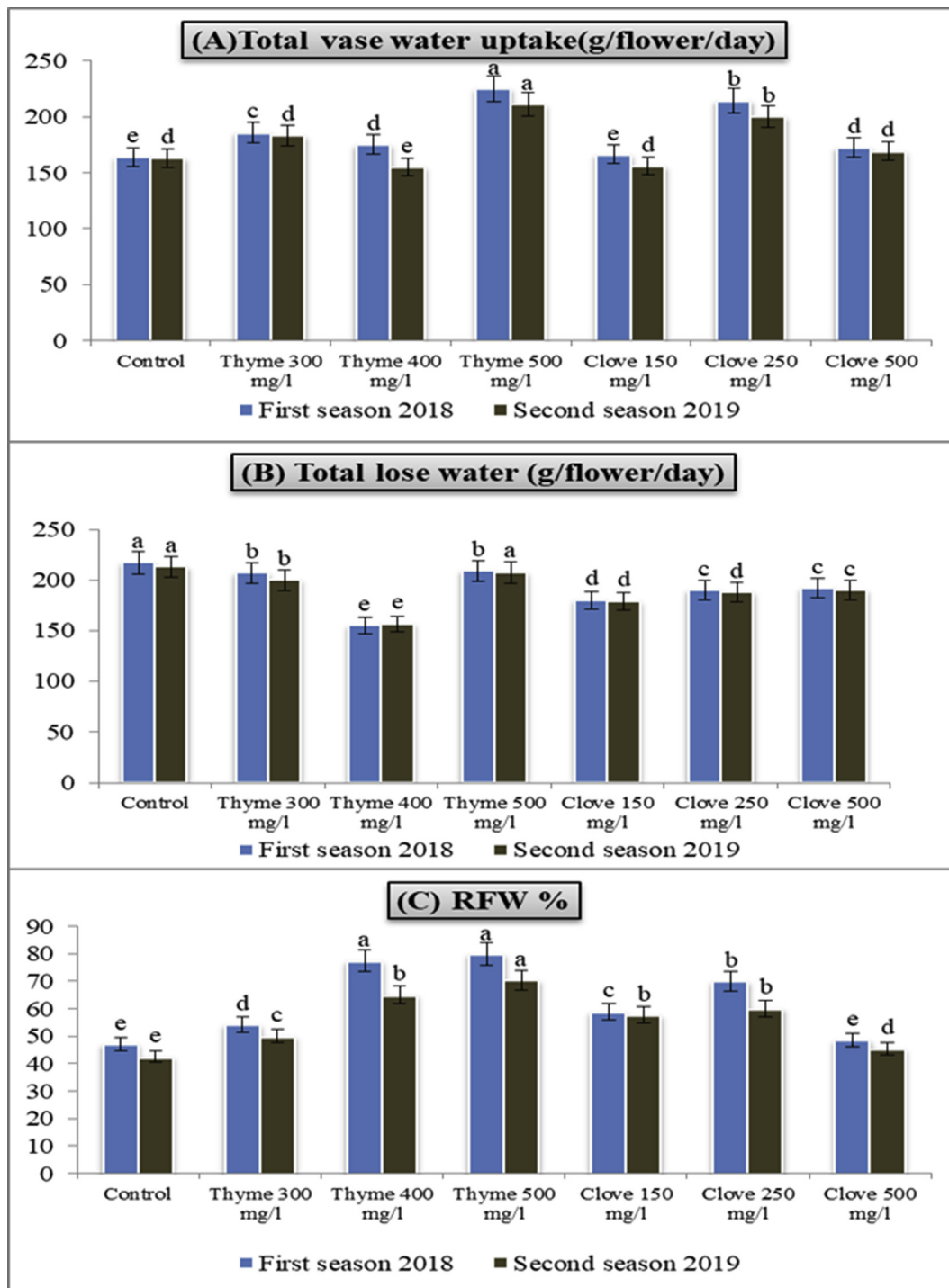


Figure 2. (A, B, and C) Effect of different concentrations of thyme and clove essential oils on water relation and relative fresh weight during 2018–2019 seasons. Columns data with different letter/s differ significantly ($p \leq 0.05$). Letters (a–e) are comparison of individual treatment means.

an increase in the vase life of cut flowers (Sharififar et al., 2007; Solgi et al., 2009; Solgi and Ghorbanpour, 2014).

3.2. Diameter of head flower and stem

The effect of the different concentrations of thyme and clove essential oils as preservatives in holding (vase) solutions on the diameter of head flowers and stem were determined individually, and the data are shown in Figure (1). The diameters of both head flowers and stem were

significantly increased with the groups treated with 500 mg/l thyme oil and 250 mg/l clove oil dissolved in preserved solution during the two tested seasons compared with the control (distilled water). The least means of the diameters of both flower head and stem were found on the preservative solution fortified with 500 mg/l clove oil. In contrast, the highest means of the diameters of both flowers head and stem was observed on the vase solution containing 500 mg/l of thyme oil compared with the control and the other concentrations of thyme and clove oils. These findings are in the same line with the study of Hashemi

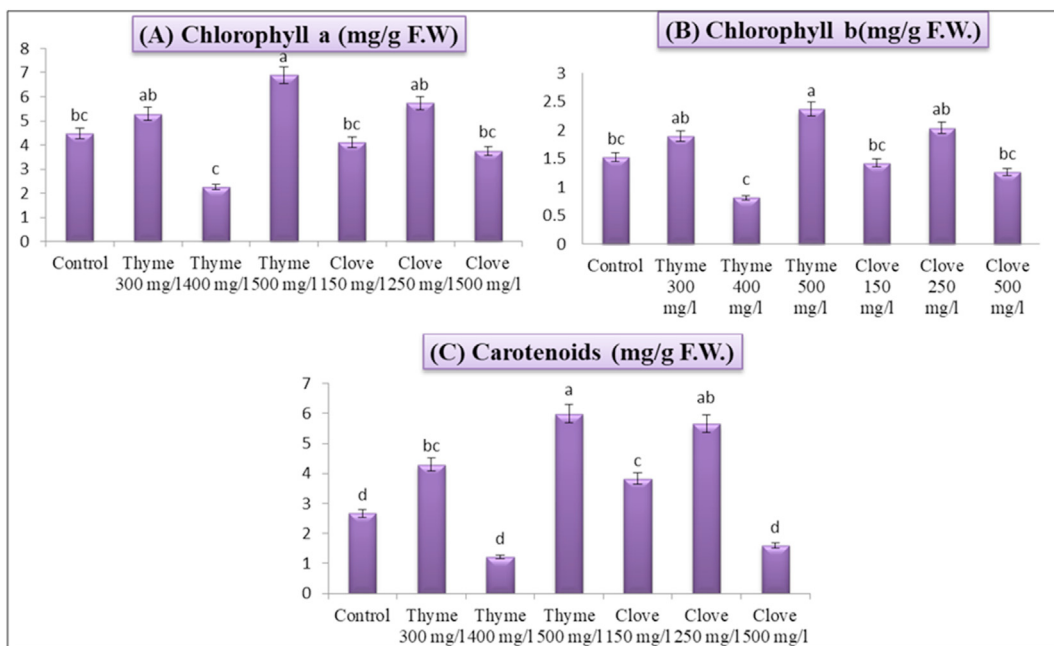


Figure 3. (A, B and C) Effect of different treatments on chlorophyll a, chlorophyll b and carotenoids content of chrysanthemum cut flowers. Columns data with different letter/s differ significantly ($p \leq 0.05$). letters (a–d) are comparison of individual treatment means.

et al. (2013), who demonstrated that the preserved solution containing thymol at 75 and 125 mg/l increased the flower head diameter of cut chrysanthemum. It may explain our result as one of the important phenolic compounds found in thyme oil is thymol.

3.3. Dry matter of flowers

Data in Figure (1) revealed that the heaviest values were found in the groups treated with 500 mg/l thyme oil in the first season and 250 mg/l clove oil in the second season as (8.66 g and 7.01 g, respectively) compared with the control treatment (5.66 g in the first season and 4.33 g in the second season one). Alternatively, the lightest mean value was found when treated preservative solution with 150 mg/l clove oil in two seasons (6.00 g and 5.66 g, respectively) compared to the control and the

other treatments. Cut flower vase life was positively correlated with dry weight. Similar results were obtained by Hegazi and EL Kot (2009), who reported the significant effect of essential oils (natural preservatives in keeping solutions) on increase the dry matter of gladiolus cut flowers.

3.4. Total vase solution uptake (g/flower)

It is quite clear from the data in Figure (2) that total vase water uptake by chrysanthemum cut flowers for all treated groups with thyme and clove oils separately was significantly increased compared to the untreated group (control), which showed a dramatic reduction and resulted in the least total vase water uptake of 164.04 and 163.05 g/flower, in the first and second season, respectively. In addition, the highest values of total vase solution uptake were recorded with the vase solution fortified

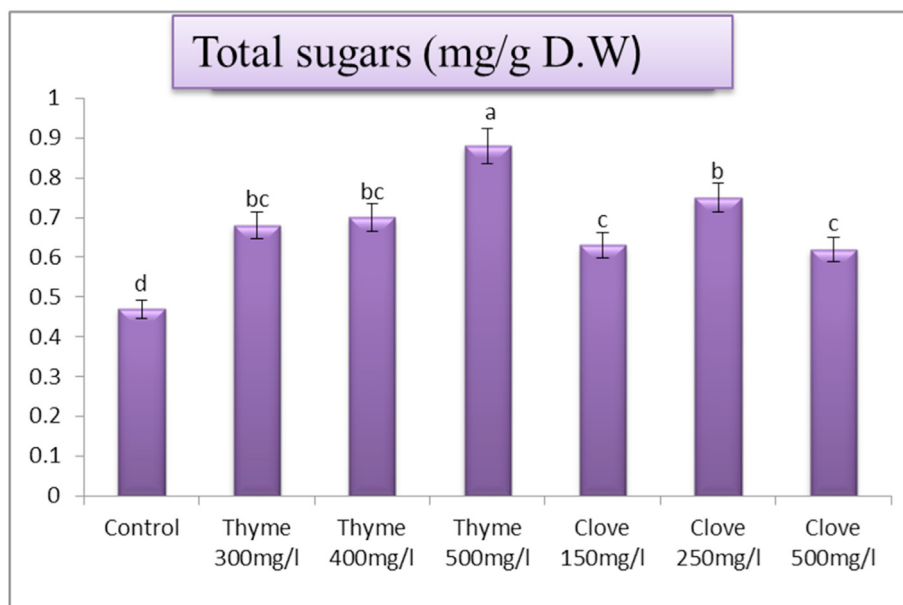


Figure 4. Effect of different treatments on total sugars (mg/gm D.W.) of chrysanthemum cut flowers. Columns data with different letter/s differ significantly ($p \leq 0.05$). letters (a–d) are comparison of individual treatment means.

with 500 mg/l thyme oil as 225.00 and 211.05 g/flower in the two seasons, respectively. Similarly, the vase solution containing 250 mg/l clove oil was significantly increased the total vase water uptake in the first and second seasons (214.40 and 200.10 g/flower, respectively). In contrast, the vase water uptake fortified with 150 mg/l clove oil showed an insignificantly increase in vase solution uptake to the untreated group.

Similar results have reported the positive effect of herbal essential oils on water uptake of cut flowers of astroemeria (Bazaz and Tehranifar, 2011). Also, Solgi et al. (2009) they indicated that the antifungal and antibacterial agents such as carvacrol, thymol, zataria oil, and thyme oil in combination with 6% sucrose had a significant impact on the shelf-life and relative solution uptake of gerbera flowers (*Gerbera jamesonii* cv. Dune'). Bayat et al. (2013) mentioned a positive response of essential oil added to the vase solution for cut flower water absorption. Similarly, Bayat et al. (2011) observed that the highest mean value of relative fresh weight in treatments including EOs compared to the control in the 6th day indicates a higher rate of water uptake by flower and a higher degree of freshness.

The vase water uptake by the cut flowers is positively related to its vase life. Therefore, the increase in the vase water uptake increases in tissue water content; it could be helpful in the hydrolysis of sugars in cells of petals through producing energy and respiration needed to improve flowers (Sakr, 2016).

Moreover, water balance achieved by the absorption of water and transpiration from cut flowers is an essential factor that affects cut flower's quality and vase life (Da Silva, 2003). So, the cut flower is subjected to water deficit when transpiration is higher than water uptake, resulting in the flower's wilting (Halevy and Mayak, 1981). The inability to absorb water is one of the causes of wilting, which may result from the growth of the microorganisms in the stem's cambial tissues (He et al., 2006).

3.5. Total water loss (g/flower)

The data in Figure (2) presented that all treated groups showed a reduction in the level of total water loss compared to the control (distilled water), with significant differences among almost all the tested treatments and the control. The minimum water loss level was observed in 400 mg/l thyme oil, followed by 150 mg/l of clove oil. In contrast, the maximum level of water loss was found in the untreated treatment in

both seasons. This result may explain as the thyme oil inhibited the ethylene producing enzymes (Babarabie et al., 2015).

3.6. Relative fresh weight (RFW %)

Data in Figure (2) indicated that most of the vase solutions used significantly gave the greatest RFW% of chrysanthemum cut flowers compared with the control (distilled water), which had the lowest RFW% of cut flowers in two seasons (47.14 and 42.45%, respectively). Whereas, the heavier values of RFW% of cut flowers recorded in the vase solution containing 500 mg/l thyme oil on both seasons (79.89 and 70.37 %, respectively) followed by the cut flowers kept in a preserved solution containing 400 mg/l thyme oil (77.41 and 65.02 %, respectively) compared to the control (distilled water) and the other concentrations of EOs tested in the preserved solutions.

Our results are agreed with the previous findings by Solgi et al. (2009), who revealed that the thymol, zataria, and thyme oils as antimicrobial agents in combination with 6% sucrose had a significant influence on the shelf-life and relatively fresh weight of the cut flowers. They established that the addition of 100 mg/l of either thymol or zataria and thyme oils positively enhanced the shelf-life to 6–7.5 days of gerbera cut flowers. Additionally, Bayat et al. (2013) reported a significant response of essential oil added with relative fresh weight and freshness of the flower.

Reducing the fresh weight of cut flowers is one of the phases of flower senescence; the more the flowers advice into senescence, the less their ability to absorb water becomes. In the end, a drastic reduction in cellular occurs (Ichimura et al., 2002). Therefore, measuring water uptake after flower harvest is one of the most significant factors of flower durability (Jowkar, 2006). Alternatively, the reduction of relative flower weight (flower relative weightless) may occur due to insufficiency of solution absorption or the rise of water loss (Bielecki et al., 1992).

3.7. Pigments content

Data presented in Figure 3 (A, B, and C) showed the effect of thyme and clove essential oils on the content of chlorophyll a, chlorophyll b, and carotenoids pigments of chrysanthemum cut flowers. The chlorophyll a, chlorophyll b, and carotenoids of chrysanthemum recorded the highest concentrations in the groups treated with 500 mg/l thyme dissolved in the keeping solution (6.89, 2.37, and 5.99 mg/g FW, respectively) compared to the control and other treatments. Similarly, the contents of

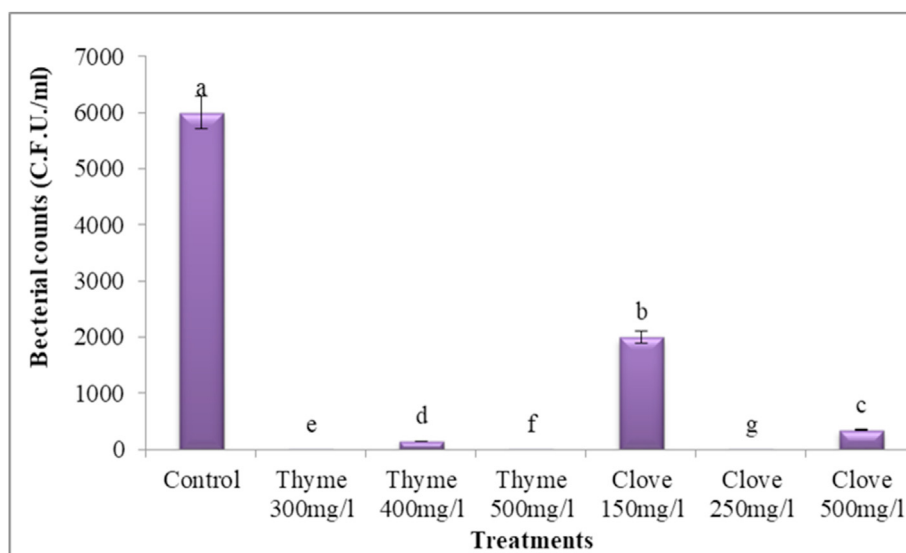


Figure 5. Effect of the preservative solution treatments on bacterial counts (C.F.U./ml) of chrysanthemum cut flowers. Columns data with different letter/s differ significantly ($p \leq 0.05$). Letters (a–g) are comparison of individual treatment means.

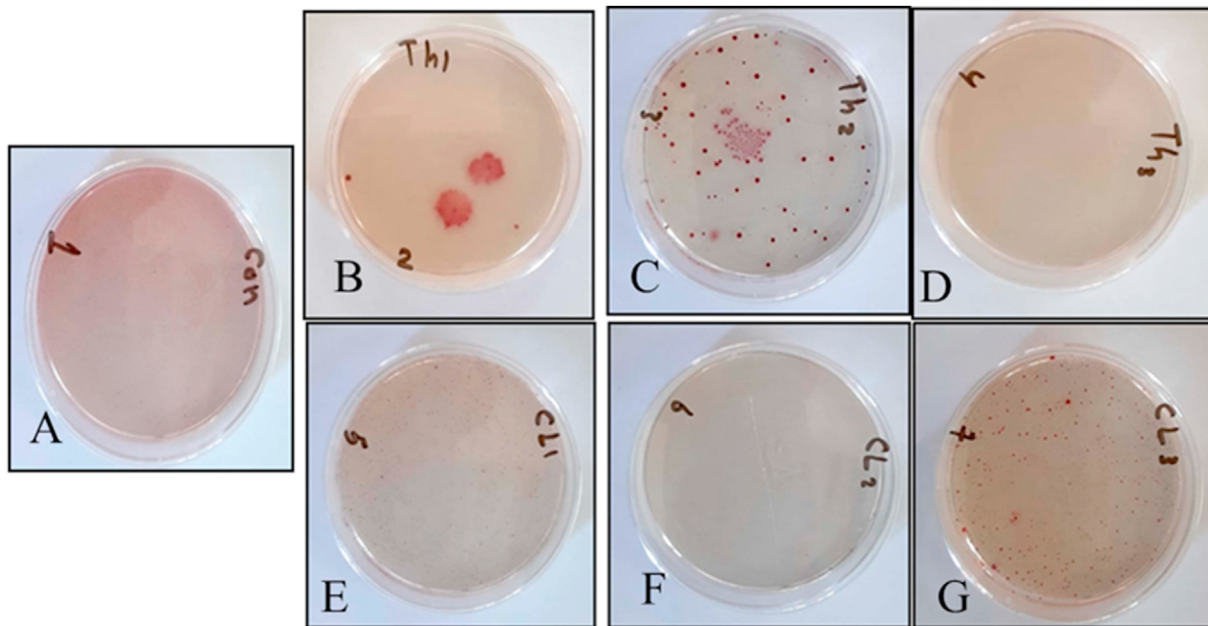


Figure 6. Effect of the preservative solution treatments on bacterial counts (C.F.U./ml) of chrysanthemum cut flowers. A: Control (distilled water) B: Thyme (300 mg/l) C: Thyme (400 mg/l) D: Thyme (500 mg/l) E: Clove (150 mg/l) F: Clove (250 mg/l) G: Clove (500 mg/l).

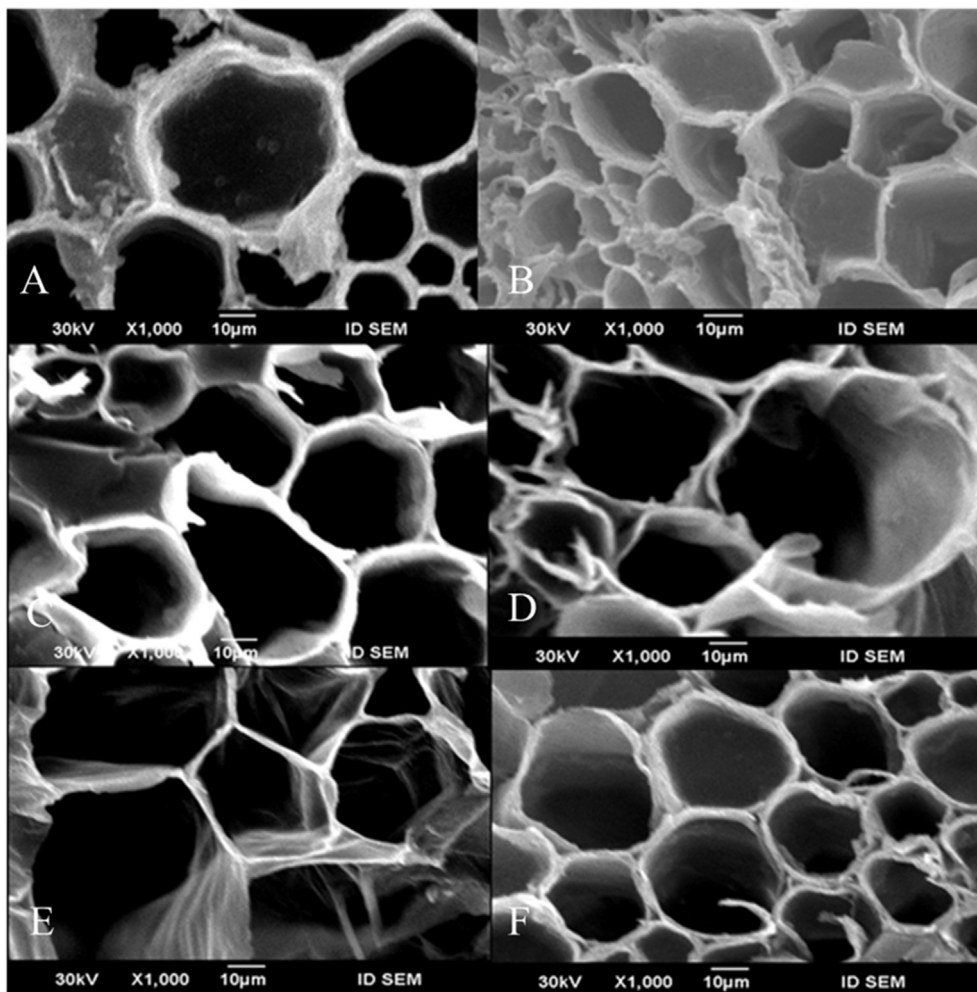


Figure 7. Scanning electron microscope micrographs of chrysanthemum xylem vessels in cross section at the base of stem show the influence of some treatments on bacterial proliferation and blockages in xylem vessels. A and B: Control (distilled water); C and D: Thyme (500 mg/l); E and F: Clove (250 mg/l).

the tested pigments were significantly increased in cut flowers treated with 250 mg/l clove oil treated groups. Oppositely, the lowest contents of chlorophyll a, chlorophyll b, and carotenoids were obtained with the vase solution containing 400 mg/l thyme oil, which gave 2.25, 0.81, and 1.21 mg/g FW, respectively.

These results in line with Massoud et al. (2015), Bazaz et al. (2015), and Hashemi et al. (2013) stated that treating chrysanthemum with essential oils increased the total chlorophyll content. Lise et al. (2004) reported that the increase in chlorophyll pigments increases cell activity and increases sugar. Many researchers also explained the maintenance or elicitation of chlorophyll when the essential oils dissolved in a vase solution because of the antioxidant properties of these oils (Babarabi et al., 2016). The water stress and obstruction of vessels increase oxygen free radicals in chloroplasts. Kazemi et al. (2014). So, Kazemi et al. (2014) demonstrated that the herbal essential oils maintained or increased the chlorophyll in lisianthus cut flowers due to their anti-radical property.

3.8. Total sugar (mg/gm D.W)

Date in Figure (4) revealed that thyme oil at 500 mg/l had a maximum value of total sugar content in dry leaves was (0.88 mg/gm D.W.), compared with the control and other treatments followed by clove oil at 250 mg/l recorded (0.75 mg/gm D.W.). In contrast, the control treatment (distilled water) recorded the minimum values of total sugar content (0.47 mg/gm D.W.).

These results are in agreement with Hashemabadi et al. (2015) mentioned that the significant positive effect of essential oil might be due to enhancing hydraulic conductivity, preventing vascular occlusion, and improving water relations. Besides, essential oils are natural antimicrobial compounds that have synergic effects on maintaining carbohydrates.

3.9. Bacterial counts (C.F.U./ml)

The data are shown in Figures 5 and 6 revealed that all treatments pulsing vase solution significantly reduced the bacterial counts (CFU/ml) of chrysanthemum cut flowers as compared with the control treatment (distilled water), which recorded the greatest total count of the bacterial cells of 6×10^3 C.F.U./ml. Furthermore, the vase solution containing 500 mg/l thyme oil and 250 mg/l clove oil had the minimum average of the bacterial count of <1 C.F.U./ml, followed by 300 mg/l thyme oil with a bacterial count of 3 C.F.U./ml compared to the other tested treatments and control.

These findings were confirmed by Solgi et al. (2009), who indicated that the antimicrobial agents such as thymol, carvacrol, zataria oil, and thyme oil in a mixture with 6% sucrose had a significant impact on the shelf- life and relative solution uptake gerbera (*Gerbera jamesonii* cv. Dune') flowers. In another study, Abo Ei-Maati (2016) found that using clove oil has an effective antioxidant and anti-microbial.

Many studies cleared that the essential oils extracted from parts of the plant have strong antimicrobial properties against many pathogens. They explained that because of the occurrence of high levels of phenolic, aldehyde, terpenes, alcohols, and flavonoids compounds such as methyl cinnamate, eugenol, e-cinnamaldehyde, alpha-pinene, citral-a, citral-b, beta-pinene, patchouli alcohol, azadirachtin, 1-8-cineole, pongamicin, and karanjin (Prabuseenivasan et al., 2006; Damjanović-Vratnica et al., 2011; Khan and Ahmad, 2011; Lavanya and Brahmprakash, 2011; Assiri et al., 2016). In addition, many studies on cut flowers were reported that the vascular blockage occurred by bacteria and other microorganisms could decrease water uptake and finally result in stem breaking and petal wilting (Vandoorn and De Witte, 1994; Nair et al., 2003).

3.10. Scanning electron microscopy (SEM)

By the Scanning electron microscopy, Figure 7 was shown that the different concentrations tested of essential oils had a positive impact in

extending the shelf-life of chrysanthemum cut flowers compared with the control (distilled water). The blockage of xylem led to water stress, which recorded the limiting factor of cut flowers' prolonging and expression as early wilting of flowers and leaves. The cross-section in Figure 5 A and B indicates the xylem cells in the control (distilled water) neighborhood of cut were filled with bacteria. As a result of this blockage, the cut flowers would lose their turgidity and decrease the chrysanthemum's vase-life cut flowers. In contrast, the treatments with 500 mg/l thyme oil and 250 mg/l clove oils show a prime state of xylem vessels.

An important antimicrobial property of essential oils and their components is their hydrophobicity, which enables them to partition with the lipids of the bacterial cell wall, cell membrane, and mitochondria, causing increased permeability of these membranes. Additionally, ions leakage from these membranes can lead to bacterial cell damage and death (Solorzano-Santos and Miranda-Novales, 2012). The bacterial populations in vase solutions of Lisianthus flowers treated with thyme oil were less than the other treatments (Kazemi et al., 2011).

Therefore, the antimicrobial activities of essential oils of herbs may be correlated with the increase chrysanthemum cut flowers' vase life. These effects have been reported by Serrano et al. (2005), who indicated that the fortification of eugenol, thymol, menthol, and eucalyptol in the storage of sweet cherries significantly reduced the growth of microbial agents. Similarly, Chanjirakul et al. (2008) reported that the use of natural compounds in some products; protects the cell's physical structure against oxidative damage caused by reactive oxygen species by increasing the antioxidant capacity of the products. Hence, cell membranes, which are the main target of reactive oxygen species, are also protected in this way.

Reactive oxygen species have a high tendency to attack cell membranes. It could be deduced that the reduction of membrane stability could most probably be associated with increases in the activity of reactive oxygen species and reduction in the antioxidant enzyme activity (Ezhilmathi et al., 2007; Chanjirakul et al., 2008).

4. Conclusion

As a final summing up, the pulse treatment by thyme and clove oils positively increased the vase life period of cut chrysanthemum cv. "Arctic Queen White" compared with the control. The best effective treatments were thyme oil (500 mg/l) and clove oil (250 mg/l), which longevity the vase life period and improved many postharvest characteristics of cut flowers such as water relation and relative fresh weight, as well as increase the contents of chlorophyll a, b and carotenoids. Additionally, the tested oils have an anti-bacterial effect. Thus, applying thyme and clove oils as natural and non-toxic essential oils in the preservative solutions showed promoting prospects for further application in prolonging the vase life of cut flowers.

Declarations

Author contribution statement

Iman Mohamed El-Sayed: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Rasha Ahmed El-Ziat: Conceived and designed the experiments; Performed the experiments; Contributed reagents, materials, analysis tools or data; Wrote the paper.

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Data availability statement

Data will be made available on request.

Declaration of interests statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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