KSÜ Tarım ve Doğa Derg 29 (Ek Sayı 1), 77-91, 2026
KSU J. Agric Nat 29 (Suppl 1), 77-91, 2026
https://doi.org/10.18016/ksutarimdoga.vi.1791262

Dose-Dependent Effects of Sewage Sludge on *Matthiola incana*: A Sustainable Approach for Ornamental Plant Production

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ABSTRACT

In recent years, the storage and disposal of sewage sludge, which have emerged as a source of pollution, have become a serious problem. In this context, the use of sewage sludge for agricultural purposes is seen as the most economical, practical, sustainable and environmentally friendly method. This study was conducted to evaluate the use of sewage sludge as a growing medium and its impact on the growth and development of *Matthiola incana* ('İron Rose Pink'). The study was conducted in 2025 in an unheated glass greenhouse belonging to the Research and Application Center of Tokat Gaziosmanpaşa University, with three replicates according to a completely randomized design. As the cultivation medium; five different media were used, 25%, 50%, 75%, and 100% sewage sludge, and a control medium (70:30 peat + perlite). The data obtained showed that sewage sludge applications at different doses showed a significant difference of 5% in plant height, while a significance level of 1% was detected in parameters such as stem diameter, raceme length, number of flowers, flower canopy width, number of leaves, leaf length and width, root length, fresh and dry weights of stem and roots, chlorophyll index, and flower color. The application of 25% sewage sludge to the growing medium yielded the best results in terms of flower canopy width and also produced high stem diameter. Applying 50% sewage sludge enhanced flower color intensity, while applying 75% sewage sludge produced the best results in terms of stem diameter and leaf development. However, the control group performed best in terms of plant height, raceme length, and number of flowers. These results indicate that sewage sludge can be used as a partial alternative to commercial growing media (peat + perlite) in the cultivation of *M. incana*; however, the application rate must be optimized according to the desired plant characteristics (e.g., plant height, flower color, leaf development).

Horticulture

Research Article

Article History
Received : 25.09.2025
Accepted : 26.03.2026

Keywords
Cut flower
Matthiola incana
Organic wastes
Sewage sludge
Sustainable agriculture

Atıksu Arıtma Çamurunun *Matthiola incana* Üzerindeki Doza Bağımlı Etkileri: Süs Bitkisi Üretimi için Sürdürülebilir Bir Yaklaşım

ÖZET

Son yıllarda, kirlilik kaynağı olarak ortaya çıkan atıksu arıtma çamurunun depolanması ve bertarafı ciddi bir sorun haline gelmiştir. Bu bağlamda atıksu arıtma çamurunun tarımsal amaçlı kullanımı ekonomik, pratik, sürdürülebilir ve çevre dostu yöntem olarak görülmektedir. Bu çalışma, atıksu çamurunun yetiştirme ortamı olarak kullanımının *Matthiola incana* ('Iron Rose Pink') bitkisinin büyüme ve gelişimi üzerindeki etkilerini incelemek amacıyla yürütülmüştür. Çalışma, 2025 yılında Tokat Gaziosmanpaşa Üniversitesi Araştırma ve Uygulama Merkezi'ne ait ısıtmasız cam serada, tesadüf parselleri deneme desenine göre 3 tekerrürlü olarak gerçekleştirilmiştir. Yetiştirme ortamı olarak: %25, %50, %75 ve %100 atıksu arıtma çamuru ile kontrol ortamı (70:30 torf + perlit) olmak üzere beş farklı ortam kullanılmıştır. Elde edilen verilerde, farklı

Bahçe Bitkileri

Araştırma Makalesi

Makale Tarihçesi
Geliş Tarihi : 25.09.2025
Kabul Tarihi : 26.03.2026

Anahtar Kelimeler
Atıksu arıtma çamuru
Kesme çiçek
Matthiola incana
Organik atık
Sürdürülebilir tarım

dozlardaki atıksu arıtma çamuru uygulamaları; bitki boyunda %5 düzeyinde önemli, gövde çapı, başak uzunluğu, çiçek sayısı, çiçek kanopi genişliği, yaprak sayısı, yaprak boyu ve eni, kök uzunluğu, gövde ve kök taze-kuru ağırlıkları, klorofil indeksi ve çiçek rengi gibi parametrelerde ise %1 düzeyinde önemli olduğu tespit edilmiştir. Yetiştirme ortamına %25 oranında uygulanan atıksu arıtma çamuru; çiçek kanopi genişliği bakımından en iyi sonuçları vermiş ve aynı zamanda yüksek gövde çapı sağlamıştır. %50 atıksu çamuru uygulaması çiçek rengi canlılığını artırırken, %75 atıksu arıtma çamuru uygulaması ise gövde çapı ve yaprak gelişiminde en iyi sonuçları göstermiştir. Bitki boyu, başak uzunluğu ve çiçek sayısında ise kontrol grubu en başarılı bulunmuştur. Bu sonuçlar, *M. incana* yetiştiriciliğinde atıksu arıtma çamurunun ticari yetiştirme ortamlarına (torf + perlit) kısmi bir alternatif olarak kullanılabileceğini, ancak uygulama oranının hedeflenen bitki özelliklerine (örneğin bitki boyu, çiçek rengi, yaprak gelişimi) göre optimize edilmesi gerektiğini ortaya koymaktadır.

Atıf İçin : Erdoğdu, S., & Kartal, H. (2026). Atıksu Arıtma Çamurunun Matthiola incana Üzerindeki Doza Bağımlı Etkileri: Süs Bitkisi Üretimi için Sürdürülebilir Bir Yaklaşım. *KSÜ Tarım ve Doğa Derg 29* (Ek Sayı 1), 77-91. https://doi.org/10.18016/ksutarimdoga.vi.1791262.

To Cite: Erdoğdu, S., & Kartal, H. (2026).Dose-Dependent Effects of Sewage Sludge on Matthiola incana: A Sustainable Approach for Ornamental Plant Production. *KSU J. Agric Nat 29* (Ek Sayı 1), 77-91. . https://doi.org/10.18016/ksutarimdoga.vi.1791262.

INTRODUCTION

Today, increasing urbanization, rapid population growth, and industrialization are leading to a rise in the amount of sewage sludge generated daily. The growing urban population, particularly from rural to urban migration, along with industrial and technological developments, has led to a rapid increase in waste generation in cities (Karacocuk et al., 2022; Işınkaralar et al., 2024). In recent years, the storage and disposal of sewage sludge, which has emerged as a source of pollution, has become a significant problem. To eliminate these problems, it is necessary to develop economical and environmentally friendly technologies for recycling waste (Topal & Topal, 2013). Recycling reusable waste with high organic content minimizes environmental impact and the amount of waste sent to landfills while also conserving resources (Çetinkale Demirkan et al., 2017). In this context, sludge from sewage sludge plants is rich in nutrients and contains important macronutrients (nitrogen, phosphorus, and potassium) when considered for use as agricultural fertilizer or soil conditioner (Soumare, 2003; Bacalu-Rus et al., 2025) and helps regulate the physical, chemical, and biological properties of soil (Souza et al., 2022). It also has the capacity to partially or completely replace the use of substrate and fertilizer by meeting the nutrient demands of plants (Ferraz et al., 2016; Souza et al., 2019; Djandja et al., 2020). Research in this field has shown that materials classified as organic waste, when used directly or mixed into the soil at certain ratios, can meet the organic matter and plant nutrient requirements of plants (Lloret et al., 2016; Antonkiewicz et al., 2018). In addition, it was concluded that sewage sludge is a potential fertilizer that can promote plant growth in agricultural areas (Nzeve & Ikubano, 2021) and is used as a soil conditioner in low-fertility soils (Manas et al., 2009; Li et al., 2009; Katkat & Aşık, 2010). In line with this approach, in many countries, sewage sludge can be used as a germination and growth medium in agricultural areas, forest areas, nurseries, and park-garden areas (Küçükhemek et al., 2005), especially in ornamental plant cultivation, which is rapidly developing and has great economic importance (López-Mosquera et al., 2011; Çetinkale Demirkan et al., 2014; Öten et al., 2016; Pekşen & Yamaç, 2016). In their study, Akat et al. (2015) obtained the most positive results from *Limonium sinuatum* by using a 75% sewage sludge mixture in the soil. However, for long-term use of sewage sludge, a 50% dose is recommended to ensure greater plant nutrient uptake and to prevent toxic effects. Furthermore, using organic waste in this way can be assumed to contribute to reducing fertilizer costs, which is a significant expense (Kartal & Geboloğlu, 2025), and thus constitutes an ecological and economic solution for waste disposal (Illera-Vives et al., 2015; Lloret et al., 2016; Antonkiewicz et al., 2018). Depending on the source, sewage sludge may contain toxic substances, heavy metals (Keller et al., 2002), harmful microorganisms (McBride, 1995; Singh & Agrawal, 2007), and parasitic insect eggs. Additionally, it sewage sludge that increases mud addition raises the levels of heavy metal activity in the environment and the soluble salt content in water (Chen et al., 2025). Salt stress, on the other hand, disrupts ionic balance in plant tissues as an abiotic stressor, negatively affecting photosynthesis and other physiological parameters due to high accumulation of Na^+ and Cl^- (Kartal et al., 2025; Erdoğdu et al., 2026). For this reason, salinity levels must be taken into account when using sewage sludge. Direct application of solid wastes or sewage sludge can lead to the

leaching of heavy metals and organic matter into the soil and ultimately to groundwater contamination (Liew et al., 2022), and their use as fertilizer in agricultural areas can lead to heavy metal pollution in the soil and the food chain (Mutethya et al., 2017). Furthermore, they persist in the environment for extended periods and accumulate in living organisms (Savas et al., 2021; Istanbullu et al., 2023; Ghoma et al., 2023). For these reasons, various countries have regulations restricting the use of sewage sludge (Martinez et al., 2002; Shober, 2003; Dolgen et al., 2007). According to these regulations, strict inspections are mandatory during the use phase and post-use period. These inspections must comply with the limit values specified in the relevant regulatory documents. Sewage sludge determined to meet legal limit values is used as a soil conditioner, organic fertilizer (on infertile soils), and in ornamental plant cultivation (Kranert et al., 2008; Akat et al., 2013; Demirkan et al., 2014). In this process, plants with high biomass production, rapid growth, well-developed root systems, effective detoxification mechanisms against excess heavy metals, and the ability to accumulate these elements at high levels in their tissues are used (Soleimannejad et al., 2020). Indeed, plants used in phytostabilization can reduce the migration of toxic metals to other areas by accumulating pollutants in their rhizomes (Thakur et al., 2016). Phytoremediation, in general, is an established strategy that uses hyperaccumulator plants to remove pollutants and waste from soil, air, and water (Wei et al., 2021). Ornamental plants not only play an aesthetic role in landscape designs, but also positively affect human health in visual, physical, and psychological aspects (Erdoğdu & Çelik, 2024). In this context, the resistance of some ornamental plant species to environmental stressors and their phytoremediation potential are important for both landscape and sustainable production; members of the Brassicaceae family, in particular, play a key role in phytoremediation technology. Many wild species of this family are known to be heavy metal hyperaccumulators and are resistant to the toxic effects of a wide range of metals (Anjum et al., 2012). One of the important species of this family in ornamental plant cultivation is the *Matthiola incana* (Stock), which we used in our study. *M. incana*, native to the Mediterranean coast, has gained popularity as an ornamental plant throughout Europe due to its diverse flower colors and pleasant fragrance. It possesses many distinct biological characteristics, including woody stem development, cold resistance, a strong floral fragrance, a long flowering period, and a high content of linolenic acid in its seed oil (Irani & Arab, 2017; Nakatsuka & Koishi, 2018; Nuraini et al., 2020).

The hypothesis of this study is that sewage sludge applied at different concentrations will affect the vegetative and generative growth parameters of *M. incana* to varying degrees relative to the control environment, and that a specific concentration (e.g., 50%) may serve as a sustainable alternative by improving flower quality. While studies on the use of sewage sludge in various plant species exist in the literature, this is the first study to comprehensively examine the effects of different sewage sludge doses on the morphological, physiological, and particularly colorimetric (color) characteristics of the high-value cut flower variety *M. incana* 'Iron Rose Pink'. In this regard, the study aims to provide valuable contributions to both the scientific literature and the ornamental plant sector.

MATERIAL and METHOD

Material

The study was carried out in a 450 m^2 glass greenhouse (40°19'55'N 36°28'33'E) with no heating or cooling system and roof ventilation at Tokat Gaziosmanpaşa University Agricultural Research and Application Center in 2025. In the study, stabilized sewage sludge was homogeneously mixed with peat (Kekkila peat) and perlite at specific volume (v/v) ratios. The control group consisted of a 70% peat 30% perlite mixture without any sewage sludge. In this study, Kekkila-brand peat, widely used as a plant-growing medium, and perlite, widely used in agriculture, were used. The sewage sludge used in this study was obtained from the Tokat Municipality Sewage Treatment Plant. After drying, the sewage sludge was passed through a 4 mm sieve to make it ready for use. The five different growing media used in the study are given in Table 1.

Table 1. Growing media
Çizelge 1. Yetiştirme ortamları

Treatments-Uygulamalar
1. Control (70% peat + 30% perlite)
2. 25% sewage sludge + 75% peat:perlite
3. 50% sewage sludge + 50% peat:perlite
4. 75% sewage sludge + 25% peat:perlite
5. 100% sewage sludge

In the study, plants were grown in pots with dimensions of 24.5*20.3*13.7 cm, and the *M. incana* 'Early Iron Rose Pink' variety was used as plant material (Figure 1). The seedlings used in the study were obtained from Seçkin Tarım, a company operating in Yalova, and were planted in the growing media during the two true-leaf stages.

Irrigation (initial watering) was applied immediately after planting the seedlings, and Hoagland's nutrient solution was applied to nourish the plants (Hoagland & Arnon, 1950). After planting, irrigation without fertilizer was applied for 10 days. The is because the seedlings' roots require water with a lower EC during the rooting process. After the tenth day, fertilization was applied in stock tanks at an EC level of 1.5 dS/m until the beginning of flowering, with an N:P:K:Ca:Mg ratio of 2:1:3:1.5:1. Fertilization was applied with the irrigation water in all irrigations. Harvesting was performed when 3-5 flower buds on each *M. incana* stem had opened, marking the optimal stage for collection. The properties of the materials (sewage sludge, peat and perlite) used in the study are given in Tables 2, 3 and 4.

Figure 1. *M. incana* 'İron Rose Pink'
Şekil 1. M. incana 'İron Rose Pink'

Table 2. Sewage sludge properties
Çizelge 2. Atıksu arıtma çamurunun özellikleri

Organic matter (%)	pH	EC (dS/m)	Zn (mg kg^{-1})	Ca (mg kg^{-1})	P_2O_5 (kg da^{-1})	K_2O (kg da^{-1})	Mg (mg kg^{-1})
35.42	5.27	3.15	172.10	11235.00	147.60	243.7	934.50
Ni (ppm)	**Fe (mg kg^{-1})**	**Mn (mg kg^{-1})**	**Cr (ppm)**	**Pb (ppm)**	**Co (ppm)**	**Cu (ppm)**	**Cd (ppm)**
57.00	69.28	39.34	98.00	19.0	3.00	40.28	1.000000

Table 3. Properties of Kekkila peat
Çizelge 3. Kekkila torfunun özellikleri

Organic matter (%)	pH	EC (dS/m)	N (mg L^{-1})	P_2O_5 (mg L^{-1})	K_2O (mg L^{-1})	MgO (mg L^{-1})
95	5.5	2.5	140	160	180	10
SO_3 (mg L^{-1})	**Fe (mg L^{-1})**	**Mn (mg L^{-1})**	**B (mg L^{-1})**	**Zn (mg L^{-1})**	**Cu (mg L^{-1})**	**Mo (mg L^{-1})**
187	0.9	1.6	0.3	0.4	1.5	0.5

Table 4. Chemical composition of perlite
Çizelge 4. Perlitin kimyasal özellikleri

Si %	Al	K	Na	Fe
33.8	7.2	3.5	3.4	0.6

Ca	Mg	O	H_2O	Others
0.6	0.2	47.5	3.0	0.2

According to long-term temperature data for Tokat province, average daytime temperatures ranged from 19.2 °C to 27.0 °C, while average nighttime temperatures ranged from 6.7 °C to 13.3 °C (Figure 2). The climatic conditions measured in the greenhouse during the study were recorded using a data logger (HOBO U12 Temperature/Humidity/Light External Data Logger). During the experimental period, the average greenhouse internal temperature was 25.57 ± 2 °C; the highest recorded temperature was 32.23 ± 2 °C, and the lowest was 18.15 ± 1 °C. The average relative humidity inside the greenhouse was documented as 60.1 ± 2%. In the greenhouse where the study was conducted, when greenhouse temperature and light intensity were high, lime was applied to the roof to control them.

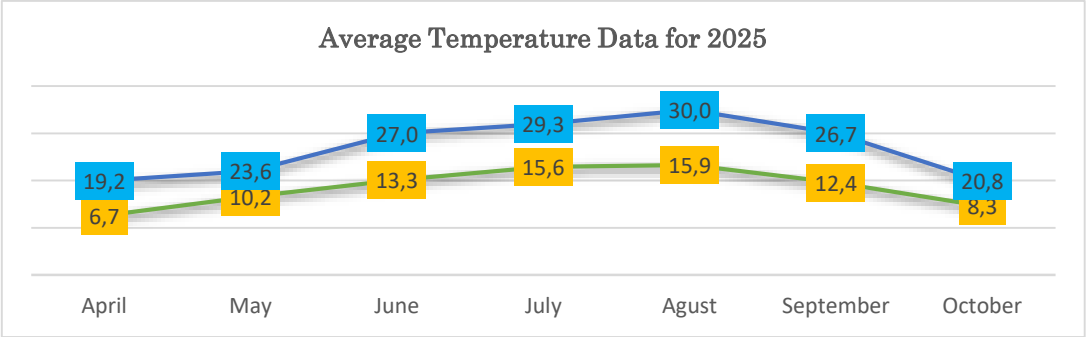

Figure 2. Monthly average minimum and maximum temperature values (°C) for Tokat Province in 2025, modeled based on long-term temperature data (MGM, 2025).

Şekil 2. 2025 yılı Tokat ili uzun yıllar sıcaklık verilerine göre modellenmiş aylık ortalama en düşük ve en yüksek sıcaklık değerleri (°C) *(MGM, 2025).*

Method

In this study, morphological and physiological parameters were investigated to determine the effects of different sewage sludge treatment rates on the growth, development, and visual quality of *M. incana* plants. Within the scope of morphological measurements; plant height (cm), number of flowers (number/plant), flower canopy width (mm), stem diameter (mm), number of leaves (number/plant), leaf width (cm), leaf length (cm), stem fresh-dry weight (g), root length (cm) and root fresh-dry weight (g) were evaluated. Chlorophyll index (SPAD value) and flower corolla color were measured as physiological parameters.

Plant height: The distance from the soil level to the tip of the shoot of each plant was measured with a ruler. Stem diameter (mm): The stem diameter of the plants was determined in mm by measuring the widest part of the stem with a digital caliper. Flower canopy width (mm): It was determined by measuring the widest part of the flower in mm using a digital caliper. Number of leaves (number/plant): It was determined by counting the number of healthy leaves on the plants. Leaf width (cm): It was determined by measuring the widest part of the leaf blade in mm using a digital caliper. Leaves were randomly taken from different parts of the plant, and the average was recorded from 3 different measurements. Leaf length (cm): The distance from the end of the leaf petiole to the tip of the leaf was determined by measuring the average in cm using a digital caliper. Leaves were randomly taken from different parts of the plants, and the average was recorded from 3 different measurements. Root length (cm): The distance from the root tip of the plants to the root collar was determined by measuring with a meter. Raceme length (cm): The distance from the beginning of the first flower row on the raceme to the flower apex was measured with a meter. Number of flowers (number/plant): The flowers on each raceme were counted and recorded. Root and stem fresh weight (g): After the plants were removed from the pots, the roots were washed with tap water, and the excess water was removed on blotting paper. Then, the fresh root and stem weights were determined using a precision scale. Root and stem dry weight (g): Roots and stems were dried in an oven at 65 °C until they reached a constant weight. The weight of the dried roots and stems was determined in g by measuring them on a precision scale (Kacar, 1972). Leaf chlorophyll content (SPAD): The amount of pigment in the leaf is important in determining the photosynthetic capacity of the plant. SPAD value in plants was measured with a SPAD device (Spad-502, Minolta, Osaka, Japan) from the fully developed middle leaves of the plant in double readings at weekly intervals for each application. Flower (Corolla) Color: L*, a* and b* values for corolla colors were determined using a Minolta CR-400. Readings were taken immediately after harvest using a Minolta device. In the Minolta device, colors, L*, illuminance, a* represents red and green color intensity, and b* represents yellow and blue color intensity. L* values range from 0 (black) to 100 (white); higher values indicate greater lightness. Positive values

indicate redness on the a* axis and yellowness on the b* axis, while negative values indicate greenness on the a* axis and blueness on the b* axis (Üren, 1999). The hue angle is defined as a color wheel with red-violet at 0° and 360°, yellow at 90°, bluish green at 180°, and blue at 270°. Chroma values indicate the saturation of a color. Chrome values decrease for dull colors and increase for vibrants colors. Hue angle (h^o) and chroma values were calculated according to equations 1 and 2 (McGuire, 1992).

$$(1)\ h^\circ = \tan^{-1}(b/a) \qquad (2)\ c = (a^2+b^2)^{1/2}$$

The study was conducted using a completely randomized design with three replications. Five plants were used per replicate, for a total of 75 plants for five different treatments. The results were analyzed using analysis of variance (ANOVA) in SPSS (IBM SPSS Statistics for Windows, Version 20.0. Armonk, NY: IBM Corp.). Duncan's multiple comparison test ($p\leq0.05$ or $p\leq0.01$) was used to determine the significance of differences between treatments.

RESULTS and DISCUSSION

In *M. incana* plants, the difference in stem diameter and raceme length between sewage sludge treatments were statistically significant at the 1% level, and plant height was significant at the 5% level. Among the different sewage sludge treatments, the best results for raceme length and stem diameter were obtained from the 25% and 75% sewage sludge treatments. Among the treatments, the best results in terms of plant height were found in the control (51.0 cm) treatment (Table 5).

Table 5. Effect of *sewage* sludge treatments on plant height, stem diameter and raceme length in the *M. incana* plant

Çizelge 5. M. incana bitkisinde atıksu arıtma çamuru uygulamalarının bitki boyu, gövde çapı ve başak uzunluğuna etkisi

Treatments Uygulamalar	Plant Height (cm) Bitki Boyu (cm)	Stem Diameter (mm) Gövde Çapı (mm)	Raceme Length (cm) Başak Uzunluğu (cm)
Control	51.0±2.00^{a}	7.76±0.34^{c}	9.71±0.60^{a}
25% sewage sludge	47.8±0.29^{b}	8.87±0.16^{ab}	8.60±0.53^{b}
50% sewage sludge	47.9±0.36^{b}	8.35±0.24^{bc}	8.00±0.58^{bc}
75% sewage sludge	48.2±1.26^{b}	9.44±0.44^{a}	8.73±0.25^{b}
100% sewage sludge	47.2±0.75^{b}	6.22±0.52^{d}	7.59±0.45^{c}
p-value	**0.015**	**0.000**	**0.004**
Level of significance	*****	******	******

******: Significant at $p\leq0.01$ level; *****: Significant at $p\leq0.05$ level

Çetinkale Demirkan et al. (2017) tested different organic wastes (fish waste, sewage sludge, and coal slag) on *M. incana* and found the raceme length to be between 12.33 and 15.89 cm. Akat et al. (2015) obtained the best results for flower number, flower stalk length, and flower stalk thickness in *Limonium* plants at 50% and above sewage sludge doses. Xue and Huang (2013) tested a mixture of sewage sludge and soil at different ratios (0%, 15%, 30%, 45%, 60%, and 75%) as a growth medium for *Paeonia suffruticosa* and reported that a mixture of 45% sewage sludge and soil produced the best plant height, flower number, and flower length. Ünal et al. (2011) investigated the effects of sewage sludge doses (0, 30, 60, 90, and 180 t ha^{-1}) on *Freesia spp.* and observed that increasing doses had positive effects on raceme and flower stalk length compared to soil. The fact that the control treatment yielded better results in plant height and raceme length can be explained by the physical properties of the sewage sludge disrupting the substrate's air-water balance and limiting plant growth, thereby affecting physiological processes. Additionally, sewage sludge yielded better results for stem diameter. This may be due to nutrients in the sewage sludge (particularly P and K) increasing cambial activity, thereby causing secondary tissue to thicken.

The differences between the number of flowers, flower canopy width, and chlorophyll index in different sewage sludge treatments in *M. incana* were found to be statistically significant at the 1% level. Among the treatments, flower number ranged from 9.00 to 16.33 number/plant, flower canopy width ranged from 24.56 to 31.01 mm, and chlorophyll index ranged from 52.33 to 60.07 SPAD. The best results were obtained from the control in terms of flower number; flower canopy width ranged from 25%, and the chlorophyll index ranged from the control and 25% sewage sludge treatment (Table 6).

Table 6. Effect of *sewage* sludge treatments on number of flowers, flower canopy width and chlorophyll index in the *M. incana* plant

Çizelge 6. M. incana bitkisinde atıksu arıtma çamuru uygulamalarının çiçek sayısı, çiçek taç genişliği ve klorofil indeksi üzerine etkisi

Treatments Uygulamalar	Number of Flowers (number/plant) Çiçek Sayısı (adet/bitki)	Flower Canopy Width (mm) Çiçek Kanopi Genişliği (mm)	Chlorophyll Index (Spad) Klorofil İndeksi (Spad)
Control	16.33±0.58^{a}	27.59±0.78^{b}	60.07±0.49^{a}
25% sewage sludge	9.70±0.58^{bc}	31.01±0.61^{a}	52.33±0.71^{a}
50% sewage sludge	10.00±1.00^{bc}	24.56±0.41^{c}	56.13±0.56^{b}
75% sewage sludge	10.70±0.58^{b}	27.84±0.96^{b}	55.73±0.68^{b}
100% sewage sludge	9.00±0.00^{c}	25.01±1.13^{c}	56.80±0.10^{c}
p-value	**0.000**	**0.000**	**0.000**
Level of significance	******	******	******

******: Significant at p≤0.01 level

Ostos et al. (2008) and Dede et al. (2009) reported that the addition of sewage sludge had a positive effect on plant growth in *Pistacia lentiscus* and *Primula vulgaris*. In their study, Patel & Patra (2014) obtained the best growth in *Tagetes mitula* from a soil-sewage sludge mixture (50:50). Similarly, Çetinkale Demirkan et al. (2014) tested different doses of sewage sludge (0, 25, 50, 75%) and found that the best results were obtained in terms of shoot length and flower number in media where 50% and 75% sewage sludge was mixed with soil. In a study on *Begonia semperflorens*, it was reported that the application of 25% sewage sludge to the medium gave the best results for flower number (Grigatti et al. 2007). Yon & Kahraman (2017) reported that different sewage materials affect the number of flowers in primroses, and that the highest number of flowers was obtained from a mixture of 50% wood chips and 50% soil after a four-week growth period. Grigatti et al. (2007) reported that the highest number of flowers was obtained from a medium containing 25% sewage sludge in *B. semperflorens*, *Mimulus spp.*, *Salvia splendens* 'Maestro,' and *Tagetes patula*. This clearly demonstrates that the response to sewage sludge varies depending on the plant species and even the variety. Furthermore, these studies show that sewage sludge ratios have different effects on flower and pedicle parameters. Hicklenton et al. (2001) determined that sewage sludge-applied media were the most suitable for plant growth performance in *Cotoneaster dammeri*, Özdemir et al. (2005) in *Cupressus macrocarpa* 'Gold Crest', and Dede (2009) in *Ligustrum, Primula*, and *Tagates*. Similarly, Wraga & Zawadzińska (2007) observed an increase in the yield and quality of chrysanthemum (*Chrysanthemum grandiflorum*) when sewage sludge was used, while Tariq et al. (2012), reported a similar result in *Dahlia hortensis* 'Figaro'. Zawadzinska & Salachna (2014) reported increased geranium (*Pelargonium sp.*) quality and yield with sewage sludge, but a negative effect at higher doses. The results showed that the control (70:30 peat:perlite) treatment yielded higher flower numbers due to the salt and nutrient imbalance in the sewage sludge. However, the presence of nutrients, including organic matter and nitrogen, in the sewage sludge increased photosynthetic capacity, resulting in noticeable differences in flower canopy width and chlorophyll index. In contrast, the higher flower canopy width observed in the 25% sewage sludge treatment may be related to the redistribution of photosynthetic products (assimilates) to different plant organs. Plants can produce different morphological outcomes by allocating their limited resources between growth and reproductive organs. In particular, studies on plant species have shown that as flower size increases, the number of flowers per individual tends to decrease (Charlesworth, 1989; Harder & Barrett 1995; Sakai & Sakai 1995; Sakai, 2000; Worley & Barrett, 2001; Sargent et al., 2007). Across species, there is a general inverse relationship between flower size and the average number of flowers per individual, indicating a balance between flower size and flower number (Kettle et al., 2011). In this context, the increase in flower canopy width despite the decrease in flower number in sewage sludge applications can be explained by the redirection of assimilates to a smaller number of flowers. Therefore, sewage sludge significantly affects generative and vegetative plant parameters, and the ratio of sewage sludge added to the medium should be carefully adjusted according to plant species.

In the study, the differences between leaf width, leaf number, and leaf length in the *M. incana* plant under different sewage sludge doses were found to be statistically significant at the 1% level. Among the treatments doses, leaf width ranged from 2.52 to 4.39 cm, leaf length from 12.53 to 17.23 cm, and leaf number ranged from 23.00 to 33.3 per plant, with the best results among all treatments being obtained from the 75% sewage sludge dose (Table 7).

Table 7. Effect of *sewage* sludge treatments on leaf width, leaf length and number of leaves in the *M. incana* plant
Çizelge 7. M. incana bitkisinde atıksu arıtma çamuru uygulamalarının yaprak eni, yaprak boyu ve yaprak sayısı üzerindeki etkisi

Treatments Uygulamalar	Leaf Width (cm) Yaprak Eni (cm)	Leaf Length (cm) Yaprak Boyu (cm)	Number of Leaves (number/plant) Yaprak Sayısı (adet/bitki)
Control	3.59±0.54^{ab}	13.21±0.59^{c}	31.7±1.53^{ab}
25% sewage sludge	3.22±0.03^{bc}	15.15±0.34^{b}	30.0±0.00^{b}
50% sewage sludge	3.70±0.76^{ab}	12.56±0.18^{c}	30.3±1.53^{b}
75% sewage sludge	4.39±0.20^{a}	17.23±0.92^{a}	33.3±1.53^{a}
100% sewage sludge	2.52±0.10^{c}	12.53±0.25^{c}	23.0±1.00^{c}
p-value	**0.004**	**0.000**	**0.000**
Level of significance	******	******	******

******: Significant at p≤0.01 level

Çetinkale Demirkan et al. (2017), in their study where they tested different organic wastes (fish waste, sewage sludge and coal slag) on wallflower plants, found the number of leaves to be between 29.67-41.0 pieces/plant. Akat et al. (2015) obtained the highest leaf number in *Limonium* plants with increasing sewage sludge doses from 50% and 75% sewage sludge. Similarly, Kahraman (2024), in their study, obtained the highest leaf width in the Toros snowdrop plant from a 25% sewage sludge medium. Tariq et al. (2012), in their study using various potting mixes on *Dahlia hortensis* 'Figaro', reported that application of sewage sludge alone and at doses above 50% had a detrimental effect on plant growth, but application doses up to 50% increased growth. A similar finding was reported by Tüfekçi et al. (2008) a, who also found that sewage sludge significantly increased the growth of *Eucalyptus camaldulensis, Eucalyptus grandis,* and *Pinus brutia* plants. In this study, the highest values for leaf width, length, and number were achieved with a 75% application rate of sewage sludge, and it was demonstrated that this dose promotes vegetative growth in the plant. This can be explained by the presence of plant nutrients in sewage sludge, particularly nitrogen, which support leaf formation and growth. Indeed, various studies have shown that nitrogen nutrition plays a significant role in regulating photosynthetic pigment synthesis in plant leaves and that the level of nitrogen supply has a significant positive correlation with the chlorophyll content of plants (Zhang et al., 2007; Zhang et al., 2013).

As a result of the study, it was observed that the differences in root length, root fresh-dry weight, and stem fresh-dry weight in the sewage sludge application doses of the *M. incana* plant were statistically significant at the 1% level. Among the treatments, the fresh root and dry stem weights were obtained from the control and 25% sewage sludge treatments, at 16.70-16.91 g and 8.50-8.60 g, respectively. The best results in root dry weight (2.84 g) and stem fresh weight (98.57 g) were obtained from the control application, while root length was determined at the 50% sewage sludge application dose (16.92 cm) (Table 8).

Table 8. Effects of *sewage* sludge treatments on root length, root fresh-dry weight and stem fresh-dry weight in the *M. incana* plant
Çizelge 8. M. incana bitkisinde atıksu arıtma çamuru uygulamalarının kök uzunluğu, kök yaş-kuru ağırlığı ve gövde yaş-kuru ağırlığı üzerindeki etkileri

Treatments Uygulamalar	Root length (cm) Kök Uzunluğu (cm)	Root Fresh Weight (g) Kök Yaş Ağırlık (g)	Root Dry Weight (g) Kök Kuru Ağır (g)	Stem Fresh Weight (g) Gövde Yaş Ağır (g)	Stem Dry Weight (g) Gövde Kuru Ağ(g)
Control	14.43±0.08^{bc}	16.91±0.93^{a}	2.84±0.03^{a}	98.57±0.53^{a}	8.60±0.36^{a}
25% sewage sludge	11.08±0.36^{d}	16.70±0.42^{a}	2.74±0.19^{ab}	89.35±0.59^{b}	8.50±0.35^{a}
50% sewage sludge	16.92±0.66^{a}	11.33±0.35^{b}	2.48±0.20^{ab}	84.16±1.50^{c}	6.70±0.25^{c}
75% sewage sludge	13.76±0.16^{c}	11.51±0.21^{b}	1.69±0.20^{c}	62.97±1.00^{e}	6.40±0.17^{c}
100% sewage sludge	15.12±0.42^{b}	11.09±0.71^{b}	2.41±0.32^{b}	65.54±0.18^{d}	7.32±0.27^{b}
p-value	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
Level of significance	******	******	******	******	******

******: Significant at p≤0.01 level

In a study in which Çetinkale Demirkan et al. (2017) tested different organic wastes (fish waste, sewage sludge and coal slag) on *M. incana* plant, the root length was between 15.22-21.67 cm, root fresh weight was between 7.44-11.80 g and root dry weight was between 1.24-1.91 g and the best results were obtained from the mixture of soil and sewage sludge. Demirkan et al. (2017) conducted a study on soil, sewage sludge + soil (1:1), fish waste +

soil (1:1), and coal slag + soil (1:1) mixtures for *M. incana* 'Iron Rose Pink' culture, and the best results in root length were obtained from the mixture of 50% sewage sludge + 50% soil. Similarly, in *L. sinuatum* species, the best result in terms of root length was obtained from the application of 50% sewage sludge (Akat et al., 2015). Dai et al. (2006) applied sewage sludge to various ornamental plant species including *B. semperflorens, Ophiopogon japonicus, Buxus radicans, Ligustrum lucidum, Viburnum macrocephalum, Osmanthus fragrans* and *Loropetalum chinense* and reported that the sewage sludge medium gave better results in all plants compared to the control. In a greenhouse study, it was reported that the application of sewage sludge to *Diplotaxis erucoides* resulted in a larger root system and increased biomass (Korboulewsky et al., 2002). In studies by Ostos et al. (2008) on *Pistacia lentiscus* and by Dede et al. (2009) on *Primula vulgaris*, the addition of sewage sludge to the medium obtained the best results for plant growth and dry weight. Similarly, Grigatti et al. (2007) reported that the application of sewage sludge increased the dry weight of *B. semperflorens* with 25% sewage sludge and *Salvia splendens* with 50% sewage sludge. However, some studies reported that the growing medium had no significant effect on root length, bulb diameter, and bulb weight (Kahraman, 2024), and the effect of sewage sludge applications on root length was insignificant (Akın & Kahraman, 2018). The discrepancy between the results reported in the literature and the findings obtained in this study may be attributed to the source of the sewage sludge used, differences in plant species, or environmental and climatic conditions during the study. When studies investigating the effects of sewage sludge doses were examined, it was concluded that different doses were beneficial for different species and that different doses should be applied to each plant (Hicklenton et al., 2001; Özdemir et al., 2005; Grigatti et al., 2007).

Pigments that constitute plant secondary metabolites (anthocyanins, carotenoids, flavonoids, and chlorophyll) (Tanaka et al., 2008; Zhao & Tao, 2015) undergo significant changes during the growth and development of ornamental plants, resulting in the emergence of a multitude of colors. Environmental factors such as pH, light, and temperature also influence the emergence of these colors (Warren & Mackenzie, 2001; Tornielli et al., 2009; Faraco et al., 2014). To date, three color systems, namely RHSCC, CIELAB, and RGB, have been widely used to classify and identify colors (Soleimanipour & Chegini, 2020; Tong et al., 2022; Wang et al., 2023). The RHSCC system facilitates simple descriptive evaluations commonly used in field research and includes L* (lightness), a* (redness, from green to red), and b* (yellowness, from blue to yellow) values (Witt, 2007). In our study, sewage sludge application doses on *M. incana* plants caused a statistically significant difference at the 1% level in L*, chroma, and hue color content. Among the treatments, the L* value (57.18) was the highest in the control group, and the flowers exhibited lighter, brighter colors. On the other hand, in 75% and 100% sewage sludge treatments, the L* values were determined as 53.98 and 54.32, respectively, and the flower color became noticeably darker. When the chroma values were examined, it was seen that 50% sewage sludge application (39.21) provided the highest color vibrancy in flowers; It was observed that color saturation decreased in the 25% sewage sludge application (30.44), and remained at a moderate level in the control (34.62) and 100% sewage sludge treatments (33.99) (Table 9).

Table 9. Effect of *sewage* sludge treatments on color content in the *M. incana* plant
Çizelge 9. Atıksu arıtma çamuru uygulamalarının M. incana bitkisinin renk içeriğine etkisi

Treatments Uygulamalar	L*	Chroma	Hue
Control	57.18±0.16^{a}	34.62±0.36^{bc}	-7.76±0.11^{a}
25% sewage sludge	55.99±0.20^{b}	30.44±5.11^{c}	-10.44±2.07^{b}
50% sewage sludge	56.95±0.23^{a}	39.21±0.79^{a}	-9.55±0.29^{b}
75% sewage sludge	53.98±0.46^{c}	37.48±0.38^{ab}	-6.87±0.35^{a}
100% sewage sludge	54.32±0.37^{c}	33.99±0.40^{bc}	-6.21±0.25^{a}
p-value	**0.000**	**0.009**	**0.001**
Level of significance	******	******	******

******: Significant at p≤0.01 level

According to these findings, applying sewage sludge increased flower color vibrancy, whereas low and high doses reduced it. Hue values were found to be more negative in the 25% and 50% sewage sludge treatments (-10.44 and-9.55), resulting in more pronounced reddish hues in the flowers. This effect diminished at higher doses, while the control and high-dose treatments exhibited more balanced hues (Table 9).

This phenomenon can be attributed to an increase in anthocyanin pigments, which play a significant role in flower color formation; indeed, anthocyanins are important secondary metabolites found in a wide variety of plant parts, including flowers, fruits, stems, and leaves (Liu et al., 2018). Anthocyanin biosynthesis and stability are influenced by various factors, including temperature and pH, and these factors can alter anthocyanin levels in plants (Jung

et al., 2020; Yamazaki et al., 2021). In this context, the chemical composition of sewage sludge (particularly pH and nutrients) may have influenced pigment formation and contributed to the observed color changes. Overall, the application of 50% sewage sludge increased flower color vibrancy and emphasized reddish hues. This may be due to the fact that the 50% sewage sludge application induces mild stress in the plant, thereby stimulating the synthesis of secondary metabolites (anthocyanins). The application of high sewage sludge doses (75% - 100%) resulted in darker colors and a decrease in vibrancy, indicating that sewage sludge doses have a significant effect on flower morphology and aesthetic characteristics. In addition, the ability of anthocyanins to scavenge free radicals and form chelates with metal ions (Mi et al., 2021; Ahammed & Yang, 2022) suggests that the introduction of trace elements into the environment through sewage sludge applications may have triggered anthocyanin synthesis by inducing mild stress in the plant. This can be considered a possible mechanism explaining the increase in color intensity observed, particularly at a 50% sewage sludge dose.

CONCLUSION

The amount of sewage sludge generated by sewage sludge plants is an ever-increasing source of pollution. The use of sewage sludge as a highly effective recycling method in agricultural areas is undeniable, both for preventing environmental problems and as an alternative solution. This is of great importance for both ecological and economic benefits. Today, growing crops in greenhouses under controlled conditions year-round has become possible. However, this development brings with it an increase in the amount of inputs used in agriculture and the generation of industrial waste materials, in addition to agricultural waste. In recent years, increasing emphasis has been placed on environmental protection and the sustainability of natural resources. This will contribute to the reuse of recyclable waste in the agricultural sector, particularly in the non-food ornamental plant sector, and to reducing environmental pressure.

This study demonstrated that applying sewage sludge at different concentrations significantly affected all growth and development parameters of *M. incana*. The control group (peat + perlite) performed best in quality parameters such as plant height and number of flowers, indicating that the use of pure sewage sludge may be limiting. However, specific sewage sludge doses of 50% and 75% outperformed the control group in key traits, including stem diameter, leaf development, and, in particular, flower color intensity. In particular, the 50% sewage sludge application's ability to enhance red tones and color vibrancy in flowers presents significant commercial potential.

In conclusion, in the cultivation of *M. incana* 'Iron Rose Pink,' mixtures containing 50% or 75% treated sewage sludge can be used as a partial alternative to peat, a scarce resource, to optimize plant growth and flower quality while reducing production costs. Future studies are recommended to investigate the underlying physiological and biochemical mechanisms (particularly the effects on pigment synthesisunderlyingof these positive effects, as well as to conduct long-term monitoring of soil health and heavy metal accumulation. Additionally, conducting similar studies with different *Matthiola* varieties and other cut flower species to determine species-specific optimal sewage sludge doses will contribute to the widespread adoption of this sustainable approach.

ACKNOWLEDGMENTS

Not applicable.

Funding

This research received no external funding.

Availability of data and materials

Data will be made available on request.

Declarations, Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Contribution Rate Statement Summary of Researchers

The authors declare equal contributions.

Conflict of Interest

All authors declare that they have no financial or other substantive conflicts of interest in their articles that could influence the results or interpretations.

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