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Eco-Friendly Novel Algicidal Efficiency of *Streptomyces eurocidicus* JXJ-0089 Against Harmful Algal Bloom of *Microcystis aeruginosa* MDEG1

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ABSTRACT

Cyanobacterial harmful algal blooms occur worldwide and cause deleterious effects on freshwater or marine ecosystems and public health. Cyanobacterial algal blooms create and release toxic odorous chemicals that are really dangerous to the biodiversity of aquatic ecosystems and the drinking water supply. The biological regulation of toxic algal blooms is essential to economic growth, protection of the environment, and promotion of human health. Treatment of the El-Manzala Lake strain of *Microcystis aeruginosa* MDEG1 (Damietta, Egypt) with *Streptomyces eurocidicus* JXJ-0089 induced an active algicidal effect and disrupted the antioxidant systems. The data revealed a significant decrease in photosynthetic pigment content (chlorophyll *a*, carotenoid, C-phycocyanin, allophycocyanin, phycoerythrin, and total phycobiliprotein), size of chloroplasts, photosynthetic efficiency, total carbohydrate, total soluble protein, and total algal biomass production. In this study, the effect of *S. eurocidicus* treatment on the physiological properties of *M. aeruginosa*, such as ROS cellular redox status using flow cytometry technique, transmission electron microscopy technologies, was estimated to investigate the ultrastructural of the cell morphology, the oxidative damage indicator malondialdehyde, and the activities of antioxidant enzymes of SOD, CAT, and POD. The main manifestations were the induction of oxidative stress, increasing ROS formation, an impaired cell membrane structure, damage to the cell membrane structure, and potentially disrupting its barrier function leading to cellular damage. These findings suggest that *S. aeruginosa* has the potential to be used as an eco-friendly biological control agent for managing harmful effects of *M. aeruginosa* cyanobacterial blooms, particularly in water treatment processes.

INTRODUCTION

The severity of global climate change has increased the magnitude, frequency, and duration of harmful algal blooms globally **(Huisman *et al.*, 2018)**. Cyanobacterial blooms are a problem for the water environment and cause deleterious effects on marine or freshwater ecosystems **(Paerl & Otten, 2013; Cai *et al.*, 2025)**. Worldwide,

cyanobacterial harmful blooms are particularly common in nutrient-rich, calm waters, and they primarily produce cyanotoxins, which are extremely harmful to the environment **(Chen *et al.*, 2011; Zhang *et al.*, 2015)**, cause the fishing and aquaculture industries to suffer enormous financial losses (**Zhang *et al.*, 2015)**, and negatively affect the health of humans **(Feng *et al.*, 2020)**. Additionally, a number of cyanobacterial species create and release odorous chemicals and poisons that endanger human health and aquatic environments **(Wang *et al.*, 2024)**. Cyanobacterial toxins are known to cause liver cancer **(Falconer *et al.*, 1983)**, skin illness **(Stewart *et al.*, 2006)**, death of humans and animals **(van Apeldoorn, 2007)**, and gastroenteritis **(Feng *et al.*, 2020)**. Furthermore, they limit the economic expansion of aquaculture, tourism, and fisheries **(Anderson *et al.*, 2012; Zhang *et al.*, 2014)**. Certain species of Microcystis produce cyclic hepatotoxic heptapeptides called microcystins, which cause health problems in aquatic organisms and terrestrial **(Yang & Kong, 2014)**.

In many regions, harmful cyanobacterial blooms have been linked to health and environmental problems like eutrophic lakes, ponds, and reservoirs, which cause serious marine environmental problems all around the world, and numerous eutrophication control techniques have been used to suppress cyanobacteria and algae in aquatic environments, including chemical algaecides, physical oxidants, allelochemicals, ultraviolet light, microwaves, clay, modified sand, plants, protozoa, Chinese traditional medicines, and cyanobactericidal microorganisms **(Kong *et al.*, 2013; Zhang *et al.*, 2014)**. Knowing that using these methods has numerous disadvantages, such as side effects, secondary pollution, high cost, limited applicability for large impoundments, and nontarget toxicity **(Lee *et al.*, 2008; Zhang *et al.*, 2015)**. Therefore, quick and extremely effective biological techniques are regarded as crucial instruments for managing cyanobacterial harmful algal blooms **(Luo *et al.*, 2013)**.

Cyanobactericidal microbial technology has garnered attention recently as a novel and safe approach to eutrophic water remediation due to its effectiveness and environmentally beneficial qualities **(Kong *et al.*, 2013)**. The ideal conditions for the development of cyanobacterial blooms include eutrophication and high temperatures **(Matthijs *et al.*, 2012)**. In nature, microorganisms that exhibit algicidal properties biologically regulate cyanobacterial harmful algal blooms. Cyanobacteria are killed by these microorganisms by attacking their cells. In order to cause direct lysis, bacteria must attach to the dangerous algae in either the direct attack or algicide release mode. In the latter case, bacteria release easily diffusible algicides, such as amino acids, proteins, or alkaloids, to destroy algal tissue **(Tang *et al.*, 2012; Zhang *et al.*, 2014)**. Proteins, amino acids, peptides, antibiotics, nitrogenous compounds, and alkaloids are among the algicidal substances secreted by various Streptomyces species **(Mayali & Azam, 2004; Sakata *et al.*, 2011)**. Previous studies suggested that the cyanobacterial or toxic algal growth inhibition may be caused by extracellular secretions from microorganisms **(Choi *et al.*, 2005; Hua *et al.*, 2009)**. These cyanobactericidal microorganisms include many

Streptomyces species **(Hua *et al.*, 2009; Tang *et al.*, 2012)**. The majority of the bioactive microbial metabolites, many of which are algicides, are produced by many Streptomyces species. These consist of triterpenoid saponin, niromycin A, nanaomycin A methyl ester, lysine, anthracidin, spiramycin, and a few unidentified proteins. **(Zhang *et al.*, 2016)**. Algicidal bacteria are crucial for controlling the growth, metabolism, and toxin generation of toxic algae **(Zhang *et al.*, 2014)**. Since algicidal bacteria suppress or lyse hazardous algae, this is thought to be the fundamental principle of microbial techniques for controlling dangerous algal blooms **(Kodama *et al.*, 2006**).

Moreover, algicide exposure would encourage cyanobacteria to release high levels of intracellular toxins and increase the level of extracellular toxins **(Ross *et al.*, 2006; Zhang *et al.*, 2015)**. Specifically, cyanobacterium blooms of *M. aeruginosa* are widespread in eutrophic lakes and reservoirs around the world **(Choi *et al.*, 2005)**. Microcystin, a hepatotoxin that affects fish, birds, livestock, wild animals, and humans, is produced as a result of cyanobacterium blooms. Microcystin is released into water bodies by cell lysis, senescence, or death (**Zhang *et al.*, 2015**).

The current study goal was to investigate the cyanobactericidal activity of *Streptomyces eurocidicus* JXJ-0089 against *Microcystis aeruginosa* MDEG1 by analyzing physiological characteristics of *M. aeruginosa*, including estimating the photosynthetic pigment (chlorophyll *a*, carotenoid, C-phycocyanin, allophycocyanin, phycoerythrin, total phycobiliprotein), size of chloroplast, photosynthetic activity, total carbohydrate content, total soluble protein, biomass production, the oxidative damage indicator of malondialdehyde, the antioxidant enzyme activities of SOD, CAT, and POD, and transmission electron microscopy for alteration in cell morphology, and the cellular redox status of reactive oxygen species were examined through utilizing the technique of flow cytometry after 14 days of treatment.

MATERIALS AND METHODS

Collection of the algal sample

The algal bloom of the *Microcystis aeruginosa* MDEG1 strain, belonging to the order Chroococcales and class Cyanophyceae, reached its highest cell density (99%) in November 2024. This strain was isolated from the brackish waters of El-Manzala Lake, Damietta, Egypt. A plankton net with a mesh size of 10μm was used to collect the algal bloom sample **(Deyab *et al.*, 2019)**. It was cultivated for five days to achieve the log phase before being employed as an inoculant in the sterile BG11 medium at 25°C and 2200 lux of white light. The light/dark cycle is 14:10 hours **(Al-Habeeb *et al.*, 2024; Helal *et al.*, 2025)**.

Cyanobactericidal bacterium

The strain of *Streptomyces eurocidicus* JXJ-0089 was purchased from China's Sinopharm Chemical Reagent Co., Ltd. The Gause's Synthetic agar medium was used to

maintain the *S. eurocidicus* culture at 5°C. The seed culture was incubated at 25°C for 72 hours while being shaken at 200 rpm to create the culture broth **(Kong *et al.*, 2013)**. The following procedures were followed prior to use of the *S. eurocidicus* fermentation broth: A filtrate free of cells was produced by filtering the mixture using a 0.25mm cellulose acetate membrane after it had been centrifuged at 13,000 xg for 15 minutes. *M. aeruginosa* culture was then inoculated with the cell-free filtrate in order to test for cyanobactericidal action **(Kong *et al.*, 2013)**.

Cyanobactericidal activity of *S. eurocidicus* against *M. aeruginosa*

The cyanobactericidal effects were investigated through the addition and dilution of 10% (v/v) *S. eurocidicus* JXJ-0089 culture broth **(Kong *et al.*, 2013)** to a 500mL conical, sterile beaker using 225mL BG11 growth culture containing *M. aeruginosa* MDEG1, delivered to a final volume of 250mL by incorporating Gause's synthetic medium **(Kong *et al.*, 2013)**. To create a negative control, 225mL of cyanobacterial solution were mixed with 25mL of Gause's medium. The previously described conditions were used for the incubation of all samples and controls. Each treatment was in triplicates ±SD.

Estimation of algal biomass

After 14 days of growth, aliquots of one liter were centrifuged at 6000 x*g* for 25min. The algal biomass following centrifugation was dried at 100°C to a stable constant weight, cooled, and weighed. The results were calculated and are presented in g L^{-1} **(Yoo *et al.*, 2010; Ratomski & Hawrot-Paw, 2021; Helal *et al.*, 2025)**.

Determination of photosynthetic pigments

Carotenoids was investigated using the technique of **Bazarnova *et al.* (2024)** and **Laylani *et al.* (2024)**. Chlorophyll *a* level in the algal medium *M. aeruginosa* was estimated according to **Moussa (2001)**, **Moussa and Khodary (2003)** and **Moussa and Hassen (2017)**. For ten minutes, the *M. aeruginosa* (V_1) was centrifuged at 5,000 rpm. After being collected, the cell pellet was reconstituted in 95% ethanol and kept for 24 hours at 5°C. After centrifuging the supernatant (V_2) for 12 minutes at 4,000 rpm, it was collected. Light absorbance was measured at 665 nm (Chl *a* absorption), 649 nm (Chl *b* absorption), and 750 nm (turbidity correction) using a spectrophotometer SP-300, Japan. Eq. (1) was used to calculate the concentration of chlorophyll *a*:

$$\text{Chl } a \text{ (mg/L)} = [(A_{665}-A_{750}) \times 13.7 - (A_{649}-A_{750}) \times 5.76] \times V_2/V_1 \quad \textbf{(1)}$$

Estimation of phycobiliprotein fractions (C-phycocyanin, allophycocyanin, phycoerythrin, and total phycobiliprotein) was carried out according to **Lamela and Márquez-Rocha (2000)**, **Moussa *et al.* (2015a)** and **Helal *et al.* (2025)**.

Estimation of photosynthetic activity

To examine whether *S. eurocidicus* affected the photosynthetic efficiency of *M. aeruginosa*, the maximum photochemical quantum yield of photosystem II (Fv/Fm) was

measured using a Phyto PAM II Phytoplankton & Photosynthesis Analyzer (WALZ, Effeltrich, Germany). After sub-sampling at the end of this experiment, a 4-mL sample was collected from each bottle and then subjected to photosynthetic efficiency measurement. Prior to the measurement, the samples were kept in the dark for 10min to relax the reaction centers of photosystem II **(Lürling *et al.*, 2018)**. The F_v/F_m ratio was measured using a saturation pulse and was determined using the following equations:

$$F_v = F_m - F_0 \quad \textbf{(1)}$$

$$F_v/F_m = (F_m - F_0) / F_m \quad \textbf{(2)}$$

Where, F_0 is the minimum fluorescence level excited by the very low-intensity measured light and F_m is the maximum fluorescence level elicited by a saturation pulse **(Park *et al.*, 2025)**. The photosynthetic efficiency was determined assuming the physiological state is represented by the highest quantum yield of PSII (F_v/F_m).

Separating the chloroplast

The process of chloroplast isolation from *M. aeruginosa* utilized the technique of chloroplast isolation buffer with 60 mM Tris–HCl, 0.35 M sorbitol, 6 mM EDTA, and pH 7.5 **(Moussa, 2011; Tirado & Combariza, 2024)**. A 50–75% Percoll gradient was used for centrifugation in order to purify crude chloroplasts. After removing the intact chloroplasts from the gradients, they were diluted three or four times and centrifuged at 3000x*g* for 4min. Subsequently, after being reconstituted in the isolation buffer, the chloroplasts were stored in the dark until they were needed again. Every operation was conducted between 0– 5°C.

Determination of chloroplast size

The dynamic technology of light scattering (Beckmann, Coulter N4 Plus apparatus, Midland, Canada) was used to determine the size distribution of *M. aeruginosa* chloroplast. The angle of scattering was equal to 90°. The mean particle size was calculated under the assumption of a unimodal distribution **(Tirado & Combariza, 2024; Helal *et al.*, 2025)**.

Estimation of antioxidant enzyme activities, malondialdehyde, total soluble protein, total carbohydrate, and ROS

According to **Dogru *et al.* (2008)** and **Kong *et al.* (2013**) the malondialdehyde level was used to measure the level of lipid peroxidation. SOD activity was measured in accordance with **Trenzado *et al.* (2006)**. POD and CAT activities were assessed in accordance with **Qian *et al.* (2009b)**. Each enzyme's activity was represented on a protein basis. **Bradford (1976)** used the bovine serum albumin as the reference standard to calculate the concentrations of total soluble proteins. Total carbohydrate content was calculated using the technique of **Singh *et al.* (2019)**. By examining 2',7'-

dichlorofluorescein's fluorescence (DCF) intensity, the ROS level was determined **(Qian *et al.*, 2012)**.

Transmission electron microscopy (TEM)

Using a Hitachi H-600 TEM, ultrathin sections stained with 5% uranyl acetate and lead citrate were examined to investigate the ultrastructural changes in *M. aeruginosa* cells treated with *S. eurocidicus* **(Kong *et al.*, 2013)**.

Statistical analysis

The statistical software (SPSS version 17, SPSS Incorporated Company, Illinois, USA) was used to conduct analytical statistics. The mean ± standard deviation (SD) of three replicates was used to present the results. To ascertain the level of significance, the collected data were statistically investigated utilizing one-way analysis of variance (ANOVA) at $P \leq 0.05$. Tukey's analysis was used and treatment means were compared at $P \leq 0.05$ **(Moussa & Galad, 2015b; Abdel-Alim *et al.*, 2023)**.

RESULTS AND DISCUSSION

Effect of *S. eurocidicus* treatment on the biomass production of *M. aeruginosa* after 14 days of growth

The growth rate of *M. aeruginosa* was severely suppressed as the viable cell percentage decreased, which in turn decreased the total biomass content after 14 days of growth by 48% as compared with the control (Table 1). Consistent with our results, **Hua *et al.* (2009)** discovered that at a 5% (v/v) treatment level, Streptomyces strain NT0401 significantly decreased the number of live *M. aeruginosa* cells. In addition to attacking cyanobacteria directly, algicidal microbes also destroy them indirectly by using amino acids, alkaloids, nitrogen compounds, proteins, peptides, antibiotics, and other substances **(Luo *et al.*, 2013)**.

Effect of *S. eurocidicus* application on photosynthetic pigment, size of chloroplast, photosynthetic activity, total carbohydrate content, and total soluble protein in *M. aeruginosa* after 14 days of growth

S. eurocidicus severely inhibited the photosynthetic pigment content of chlorophyll *a*, carotenoid, C-phycocyanin, allophycocyanin, phycoerythrin, and total phycobiliprotein in *M. aeruginosa* by 61, 52, 41, 20, 33, 50, 34, 60, 48, 37, and 59 % as compared to the control after 14 days of growth (Table 1).

The main pigments involved in photosynthesis are chlorophylls (primarily chlorophyll-*a* and chlorophyll-*b*), carotenoids, and phycobilins. These pigments are responsible for capturing light energy, which is used to drive the process of photosynthesis **(Helal *et al.*, 2025)**. The photosynthetic pigment is a crucial marker of photosynthesis and algal growth **(Cai *et al.*, 2025)**. Other recent investigations have also documented the cyanobactericidal action of microorganisms against a variety of cyanobacteria **(Kim *et al.*, 2008)**. Decreased photosynthetic efficiency is related to

reduction in the ability of the cells to perform photosynthesis, impacting their energy production. *S. eurocidicus* treatment induced a significant decrease in the size of chloroplast by a 34 % in *M. aeruginosa* as compared to the control (Table 1).

Application of *S. eurocidicus* to *M. aeruginosa* decreased significantly the total soluble protein and total carbohydrates after 14 days by 58.8 and 37.6%, respectively, in comparison with the control (Table 1).

Effect of *S. eurocidicus* on cyanobacterial activities of POD, SOD, CAT, and MDA

Activities of cellular enzymes, such as SOD, POD, and CAT, were determined to investigate the cellular defense response induced by *S. eurocidicus* stress. Table (2) shows that *S. eurocidicus* treatment induced a significant increase in the level of POD, SOD, CAT, and MDA in the algal tissue of *M. aeruginosa* by 72, 68, 67, and 79%, respectively, relative to the control after 14 days of growth.

Hong *et al.* (2008) and **Qian *et al.* (2009a)** have noted that the majority of antioxidant enzyme activity increased during stress and that the strong membrane lipid peroxidation was a crucial indicator of increased cellular damage. Unsaturated phospholipid-based cell membranes were susceptible to oxygen attack, which led to an accumulation of MDA and frequently induced membrane lipid peroxidation. Antialgal substances have the potential to severely disintegrate the cellular morphology **(Zhang *et al.*, 2015)** and SOD function, decrease the level of chlorophyll *a*, and increase ROS. This in turn causes lipid peroxidation with malondialdehyde as a breakdown product **(Feng *et al.*, 2013; Zhang *et al.*, 2015)**.

Hong *et al.* (2008, 2009) and **Qian *et al.* (2008a, b)** also observed that *S. eurocidicus* caused membrane lipid peroxidation and cell membrane damage, as demonstrated by the present study's ability to increase MDA levels and MDA content. Antioxidant enzyme activity, including that of POD, SOD, and CAT, clearly showed a considerable increase in exposure to *S. eurocidicus* **(Kong *et al.*, 2013)**. The consistent increase in CAT, SOD, and POD activities in algae suggested that these antioxidant enzyme activities may be a major site of action **(Qian *et al.*, 2008a, b)**; allelochemicals exposure also caused a severe decrease in the activities of antioxidant enzymes in cyanobacteria or algae **(Kong *et al.*, 2013)**. Three essential cell antioxidases that shield organisms form harm brought on by oxygen-free radicals are SOD, CAT, and POD **(Shabana *et al.*, 2017)**. **Mccord and Fridovich (1969)** suggested a theory that has been widely used in toxicology to explain superoxide radical damage to organisms.

The production of ROS, which include hydroxyl radicals, hydrogen peroxide, and superoxide anion radical, occurs during membrane-linked electron transfer and is found in chloroplasts, peroxisomes, and mitochondria **(Kong *et al.*, 2013)**. Furthermore, cyanobacteria and green algae produce an excess of ROS as a response to cyanobactericidal stress, which can cause oxidative damage either indirectly or directly by causing an elevated level of ROS **(Wang *et al.*, 2011)**. Thus, it was evident from the

study's sharp increase in ROS levels that *M. aeruginosa* cells were experiencing severe oxidative stress **(Kong *et al.*, 2013)**.

Furthermore, owing to the elevated ROS levels, oxidative damage occurred on proteins, nucleic acids, and membrane lipids. Other interactions between allelochemicals and cyanobacteria have shown this effect. According to a prior study, during stress, the cyanobacterium *M. aeruginosa* ROS level dramatically increased to 1.91 times that of the controls **(Kong *et al.*, 2013)**. The findings are in agreement with earlier research postulating both cellular membrane structural damage and an increase in membrane permeability **(Hong *et al.*, 2008)**.

The synthesis of Chl *a*, protein, and carbohydrates is typically linked to the growth of algal, cyanobacterial, and diatomic cells. The amounts of cellular protein and carbohydrates are two fundamental markers that reflect the physiological condition of most algae tissue **(Kong *et al.*, 2013)**. The experiment's control group's increased cellular protein content suggested that new protein was produced. Typically, proteins, lipids, nucleic acids, neutral and charged polysaccharides, and small molecules make up the cyanobacterial organic matter. Of these, polysaccharides can account for as much as 80–90% of the total release **(Kong *et al.*, 2013)**.

Table 1. Effect of *S. eurocidicus* application on chlorophyll *a*, carotenoid, C-phycocyanin, allophycocyanin, phycoerythrin, total phycobiliprotein, size of chloroplast, photosynthetic activity, total carbohydrate content, and total soluble protein in *M. aeruginosa* after 14 days of growth

Parameter	Control	*M. aeruginosa*
Chlorophyll *a* (mg g^{-1}FW)	19.1 ± 1.1^{a}	7.4 ± 0.4^{b}
Carotenoid (mg g^{-1}FW)	65.3 ± 3.3^{a}	31.3 ± 2.2^{b}
C-phycocyanin (mg g^{-1}FW)	12.5 ± 0.9^{a}	7.4 ± 0.4^{b}
Allophycocyanin (mg g^{-1}FW)	10.9 ± 0.2^{a}	8.1 ± 0.4^{b}
Phycoerythrin (mg g^{-1}FW)	7.8 ± 0.6^{a}	5.2 ± 0.3^{b}
Total phycobiliprotein (mg g^{-1}FW)	38.1 ± 1.8^{a}	18.9 ± 1.3^{b}
Size of chloroplasts (nM)	785 ± 70.1^{a}	521 ± 45.2^{b}
Photosynthetic activity (F_v/F_m)	0.5 ± 0.02^{a}	0.2 ± 0.01^{b}
Biomass production (g L^{-1})	2.7 ± 0.13^{a}	1.4 ± 0.07^{b}
Total carbohydrate content (mg L^{-1}))	197 ± 11.8^{a}	123 ± 7.6^{b}
Total soluble protein (mg L^{-1})	85 ± 5.7^{a}	35 ± 2.2^{b}

Values are represented as the mean ± SD of triplicate. Means marked with the same superscript letters are not-significant ($P>0.05$), whereas others with different superscript letters are significant ($P<0.05$).

The TEM ultrastructural analysis showed that *M. aeruginosa* cells were damaged. This finding suggested that the photosynthetic complex disintegrated as a result of the cellular structure disappearing. According to the results of flow cytometry and TEM, the variation in carbohydrate and protein contents suggested that some organic substances were produced by the chlorotic *M. aeruginosa* tissue, and the significant increases in ROS and activities of antioxidant enzymes suggested that the breakdown of the cell and tissue structure may be the result of an imbalance in the antioxidant defense system caused by oxidative stress in *M. aeruginosa* caused by *S. eurocidicus* application. This could be a sign of the evident disintegration of the *M. aeruginosa* tissue **(Kong *et al.*, 2013)**.

Table 2. Effect of *S. eurocidicus* treatment on superoxide dismutase (SOD), peroxidase (POD), catalase (CAT), and malondialdehyde (MDA) in *M. aeruginosa* after 14 days of growth

Parameters	Control	*M. aeruginosa*
SOD (U mg^{-1} protein)	48 ± 2.4^{a}	83 ± 5.5^{b}
POD (U mg^{-1} protein)	67 ± 4.1^{a}	98 ± 8.8^{b}
CAT (U mg^{-1} protein)	112 ± 7.4^{a}	167 ± 13.1^{b}
MDA (µg L^{-1})	76 ± 4.1^{a}	96 ± 7.7^{b}

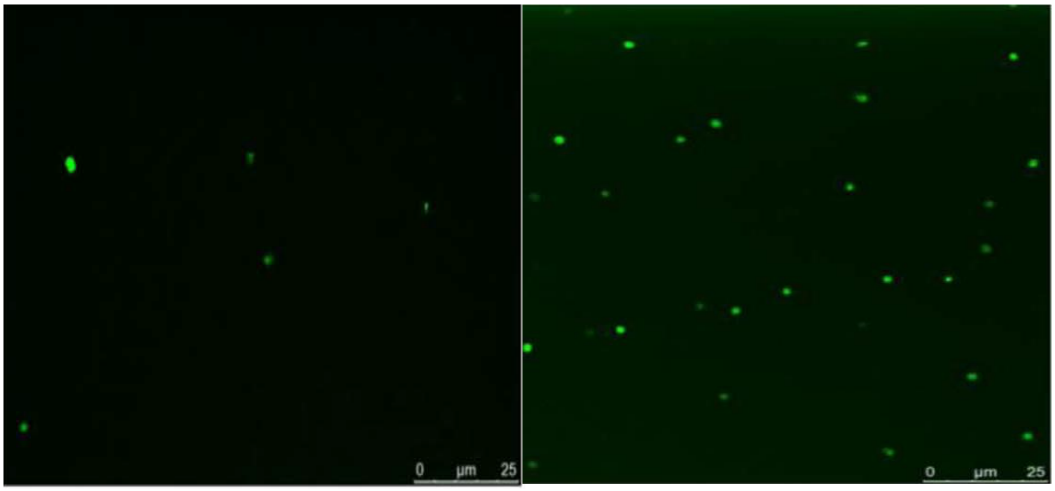
Values are represented as the mean ± SD of triplicate. Means marked with the same superscript letters are not-significant ($P>0.05$), whereas others with different superscript letters are significant ($P<0.05$).

Effect of *S. eurocidicus* on cyanobacterial ROS levels

M. aeruginosa growth suppressed by *S. eurocidicus* was measured in terms of ROS levels to determine whether the bacteria's cells were under oxidative stress. The fresh cyanobacterium exposure to *S. eurocidicus* showed weak DCF fluorescence in the control group and the fluorescence intensity dramatically increased after 14 days of growth (Fig. 1).

Numerous investigations have demonstrated that environmental stressors cause cyanobacteria to produce excessive amounts of ROS, which inhibits their ability to proliferate **(Hong *et al.*, 2009; Tang *et al.*, 2012; Mohammed *et al.*, 2020)**.

Malondialdehyde levels and antioxidant enzyme activities, such as those of catalase, superoxide dismutase, and peroxidase, were found to be significantly elevated in stressed *Chlorella vulgaris* **(Qian *et al.*, 2008b; Mohammed *et al.*, 2025a, b)**. However, the allelopathic effect of pyrogallic acid and allelochemical gramine on *M. aeruginosa* may induce severe oxidative damage and increase lipid peroxidation **(Hong *et al.*, 2009; Shao *et al.*, 2009)**.



(a) **(b)**

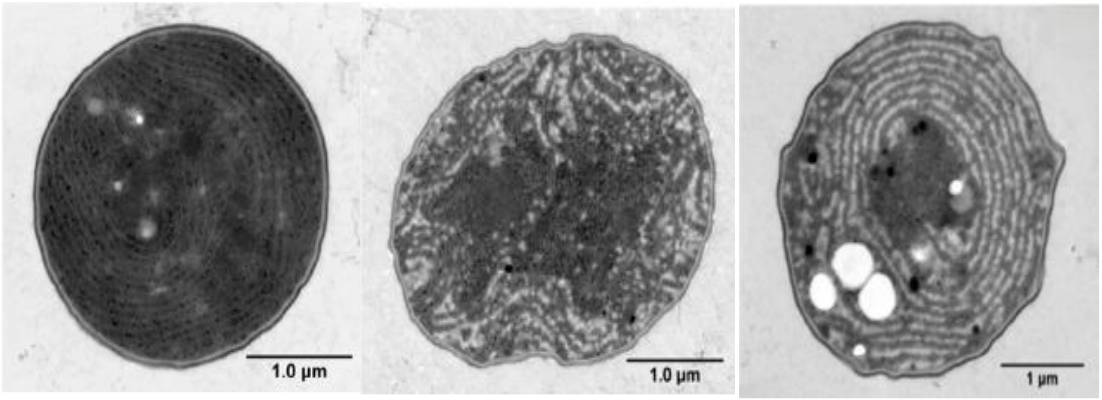
Fig. 1. The effect of *S. eurocidicus* on *M. aeruginosa* intracellular ROS production. A laser confocal microscope was used to identify the intracellular ROS. **(a)** control of *M. aeruginosa*; **(b)** *M. aeruginosa* after 14 days of growth

Effects of *S. eurocidicus* on cyanobacterial cell morphology and growth inhibition

The impact of *S. eurocidicus* on the cell morphology of *M. aeruginosa* strains was examined using TEM to analyze changes in the cell morphology after 7 and 14 days of growth (Fig. 2).

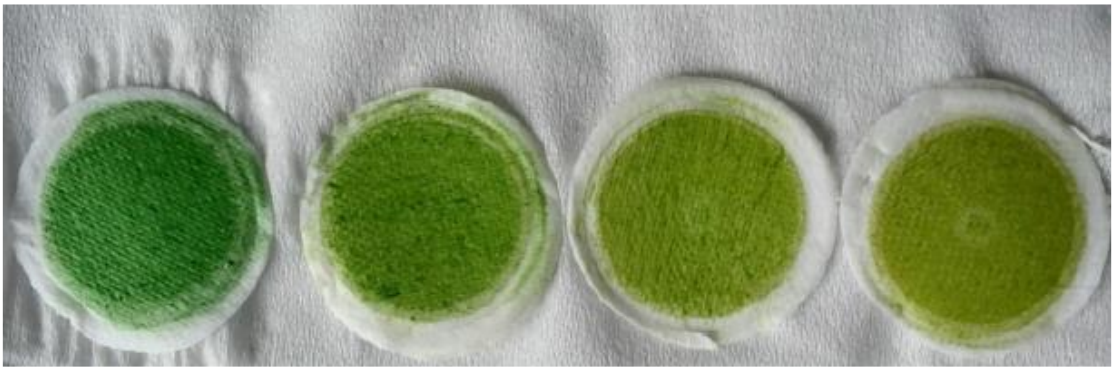
S. eurocidicus exposure for 7 days caused slight lysis of cell membranes in *M. aeruginosa* and led to minor distortion of the thylakoid (Fig. 2b). Its impact on *M. aeruginosa* strain cell morphology was more pronounced, though, as the cells changed from their typical spherical shape to an irregular oval shape, collapsed thylakoid membrane stacks, displaying crumpled cell walls, ruptured cell membranes, and the appearance of many vacuolizations on the inside (Fig. 2c). According to these results, treating *M. aeruginosa* with *S. eurocidicus* may inhibit growth and disrupt cell morphology **(Kong *et al.*, 2013)**.

Application of *S. eurocidicus* to *M. aeruginosa* algal bloom caused varying degrees of growth inhibition by decreasing the synthesis and increasing breakdown of photosynthetic pigments and decreasing its synthesis (Fig. 3). The picture indicated that the algal cells of became brown in color after 14 days of growth as comaped to the control (Fig. 3 b,c and d). These findings agree with that of **Helal *et al.* (2025)**.



(a) **(b)** **(c)**

Fig. 2. TEM micrographs showing ultrastructural changes in *M. aeruginosa* treated with *S. eurocidicus*. **(a)** control; **(b)** after 7 days; **(c)** after 14 days of growth



(a) **(b)** **(c)** **(d)**

Fig. 3. Treatment of *M. aeruginosa* with *S. eurocidicus* caused varying degrees of growth inhibition. **(a)** control; **(b)** after 5 days; **(c)** after 10 days; **(d)** after 14 days of growth, the algal cells became brown in color

CONCLUSION

The isolated strain of *Streptomyces eurocidicus* JXJ-0089 may inhibit the growth of *Microcystis aeruginosa* MDEG1: its culture broth showed remarkably strong cyanobactericidal activity. The analytical method of flow cytometry was used to assess the *M. aeruginosa* cells physiological status during the cyanobacterial inhibitory process, and the findings demonstrated that upon exposure to the cyanobactericidal culture broth, the biomass production of living cells significantly dropped. The cyanobactericidal process of *S. eurocidicus* JXJ-0089 on *M. aeruginosa* MDEG1 was found to have three

stages, according to transmission electron microscopy analysis: organelle release, cell breakage, and cell death. The increased enzyme activities of POD, SOD, and CAT activities, as well as the reactive oxygen level, suggested that strong oxidant damage and decreased membrane integrity may be the mechanism of action. Based on these findings, it is determined that *S. eurocidicus* exhibits exceptional cyanobactericidal activity by destroying *M. aeruginosa* subcellular structure in addition to influencing antioxidant enzyme activities and ROS levels.

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Author contribution
All authors participated in this research's conception, design, material preparation, data collection, and analysis, or paper draft. All authors have read and approved the final manuscript.

Declaration of Conflict of Interest
All authors declare that there is no conflict of interest.

Data availability
Contact the author for data**.**

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