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UV irradiation-promoting effect on the antibacterial activity of cyanobacterial extracts against plant pathogens: a first record

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Abstract

Background: Cyanobacteria possess a wide array of bioactive compounds including those with antimicrobial activities. The target was to investigate the UV effect on antimicrobial activity of cyanobacterial extracts. Several cyanobacterial strains were isolated from Eastern region of KSA as well as three plant pathogenic bacterial strains. Four cyanobacterial strains were used. Two strains were isolated from Al Uquair region, Arabian Gulf, and identified as *Synechococcus* sp. and *Oscillatoria* sp. The two other strains were collected from brackish stream of underground water and were characterized as *Synechocystis* sp. and *Phormidium* sp. The antimicrobial bioassay was then performed using cyanobacterial aqueous extracts. The antimicrobial effect was estimated by measuring the inhibition zone compared to that of control. The extract was divided into 2 parts: part was not exposed to UV and the other was exposed to UV-B irradiation for 10 min. The antimicrobial bioassay was performed for both parts of the extract, using plant pathogenic bacteria, namely, *Erwinia* sp., *Pseudomonas* sp., and *Bacillus* sp.

Main body: The antimicrobial profile was examined and results showed that the extracts showed non-antimicrobial effect before UV irradiation, and antimicrobial effect after UV exposure. Also, those that were active against pathogenic bacteria were more active after UV exposure. This is mostly attributed to a high optical energy of UV irradiation that subsequently had a significant impact on the electron transitions in the molecules of the extract rendering some of them more effective in their antibacterial action.

Conclusion: This short communication was the first report where the UV can alter the antimicrobial profile of cyanobacterial extracts. This is a novel approach in enhancing antibacterial activity. Future molecular investigations will be conducted to further characterize the isolate whose extract showed the highest response to UV treatment.

Keywords: Antimicrobial, Aqueous extract, Cyanobacteria, UV irradiation

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Background

UV is an electromagnetic radiation that is transmitted in waves of varying wavelengths and frequencies (Tsoulfanidis 1995). Ultraviolet radiation is mostly non-ionizing except for under certain conditions on some molecules (Kovács and Keresztes 2002). UV radiation can be divided into UV-A (315-400 mμ), UV-B (280-315 mμ), and UV-C (190-280 mμ) (Pattanaik et al. 2007). UV-A can cause lipid peroxidation, chlorophyll photo-bleaching, and inhibition of growth (Castenholz and Garcia-Pichel 2002). UV-B is also harmful but can be absorbed partly by ozone. It inhibits synthesis of chlorophyll a, nitrogen fixation, and ATP-synthase. UV-B also directly targets DNA causing dimerization between adjacent pyrimidine bases of DNA (Castenholz and Garcia-Pichel 2002). UV-C is strong but it does not reach the biosphere (Pattanaik et al. 2007). The effect of UV irradiation on cyanobacterial growth was studied (Babu et al. 1998). Nevertheless, there is still not much research on the effect of UV irradiation on the production of bioactive metabolites from cyanobacteria. In that regard, El Semary et al. (2015) demonstrated the significant effect of UV irradiation on cyanobacterial cells. UV treatment resulted in a drastic increase in the antimicrobial activity of cyanobacterial extracts. Nonetheless, the effect of UV on cyanobacterial extracts has never been studied, to the best of our knowledge. This is a novel approach in which the use of UV-B irradiation at a limited dose is tested for its ability to introduce positive change in the antimicrobial action of cyanobacterial extracts that have never been reported before. To investigate this, several extracts from marine and freshwater cyanobacterial strains were used against both gram-positive and gram-negative bacterial isolates from Al Ahsa Governorate, KSA.

This study aimed to study the effect of UV irradiation on strains from various habitats and nature.

Material and methods

Cyanobacterial extracts

Water samples were collected from marine (Al Uqair coast, Eastern Province, Al Ahsa, KSA) and freshwater habitats (canals fed from underground brackish water, Al Ahsa Governorate, KSA). Samples were examined by light microscopy, and cyanobacterial cells were isolated and cultured using F/2 for marine cyanobacteria (Guillard and Ryther 1962) and Bold's medium (Stein 1980) for brackish cyanobacteria. Cyanobacterial cultures were purified according to standard techniques and monospecific cultures were established. About 0.5 g of fresh biomass was ground in 5 ml water and centrifuged where the extract was taken and condensed to 1 ml.

Bacterial strains

Three pathogenic bacterial strains were isolated according to standard microbiological techniques from local crops. The pathogenic bacterium, *Erwinia* sp., was isolated from infected pear whereas a pathogenic strain identified as *Pseudomonas* sp. was isolated from infected tomato (Gram-negative bacteria) as well as another strain, *Bacillus* sp. (Gram-positive bacteria).

Antibacterial bioassay

Sensitivity of pathogenic strains to the extracts was assessed by using the modified Kirby Bauer Disk Diffusion Susceptibility method (Bauer et al. 1966). Whatman No. 1 sterilized-paper disks were saturated with 30 μl of extracts. Disks were dried and placed on the surface of the inoculated medium. The plates were then kept for 2 h at 4 °C to ensure the diffusion of the bioactive material, and then were incubated at 37 °C. Disks containing 30 μl of water were left to evaporate and used as negative control.

UV treatment

The cabinet radiates UVB type (280 nm) and the sample located 25 cm from the UV lamp. The exposure continued for 10 min. UVP device (California 91786).

Results and discussion

Four cyanobacterial isolates were purified and cultured. The isolates were identified using a light microscopy. Table 1 showed the phenotypic characteristics of those isolates which are from diverse habitats and exhibit morphological differences. With regard to antimicrobial bioassay, it was performed before and after exposure of cyanobacterial extracts to UV. Table 2 showed the antimicrobial profile from different cyanobacterial extracts before and after UV exposure. The extracts were ineffective against *Pseudomonas* sp. before UV exposure. However, they were effective as an antibacterial agent after exposure. Similarly, the extracts had no or little effect on the two other pathogenic bacteria. Nonetheless, they had potent antimicrobial activity after UV exposure. Negative control in all cases yielded a null (0 cm) inhibition zone.

Table 1 The cyanobacterial isolates used in the antimicrobial screening as sources of aqueous extract

Name	Habitat	Morphology
Phormidium sp.	Brackish water	Unbranched filamentous, non-heterocystous
Synechocystis sp.	Brackish water	Coccoid unicellular
Synechococcus sp.	Marine	Coccoid unicellular
Oscillatoria sp.	Marine	Filamentous, non-heterocystous, unbranched

Table 2 The inhibition zone before and after UV exposure

Bacterial pathogen	Cyanobacterial extract	Inhibition zone before exposure	Inhibition zone after exposure
Erwinia sp.	*Phormidium* sp.	**0.0 ± 0.0**	**1.1 ± 0.1**
	Synechocystis sp.	**0.9± 0.1**	**1.2 ± 0.1**
	Synechococcus sp.	**0.0 ± 0.0**	**0.9 ± 0.1**
	Oscillatoria sp.	**0.0 ± 0.0**	**0.9 ± 0.1**
Pseudomonas sp.	*Phormidium* sp.	**0.0 ± 0.0**	**1.0 ± 0.1**
	Synechocystis sp.	**0.0 ± 0.0**	**1.0 ± 0.1**
	Synechococcus sp.	**0.0 ± 0.0**	**0.8 ± 0.1**
	Oscillatoria sp.	**0.0 ± 0.0**	**1.0 ± 0.1**
Bacillus sp.	*Phormidium* sp.	**0.0 ± 0.0**	**1.1 ± 0.1**
	Synechocystis sp.	**1.0 ± 0.1**	**1.5 ± 0.1**
	Synechococcus sp.	**0.0 ± 0.0**	**0.8 ± 0.1**
	Oscillatoria sp.	**0.6 ± 0.1**	**0.9 ± 0.1**

UV radiation has mutagenic and carcinogenic effects and although it is mostly non-ionizing, it probably causes photochemical changes. It causes damage of enzymes and cellular components as it causes oxidative stress via increasing reactive oxygen (Castenholz and Garcia-Pichel 2002). Cyanobacteria, however, developed several strategies to counteract the harmful effect of UV. Cyanobacteria overcome its effect by producing specific secondary metabolites such as myosporin-like amino acid (Holzinger and Lütz 2006) and scytonemin (El Semary and Khalif 2016). The antimicrobial effect of cyanobacterial extracts may be due to many reasons including cell membrane damage of pathogenic bacteria, protein damage, and oxidative damage by increasing reactive oxygen or by DNA interference (El Semary et al. 2015). Nonetheless, no reports are available on the effect of UV on cyanobacterial extracts. The interest in extracts stem from the need to induce chemical changes without mutagenizing the original source of extracts, i.e., cyanobacterial cells thereby establishing genetically modified organisms of unpredictable impact. It is highly likely that aqueous extracts have bioactive compounds that when receiving UV can be modified further and achieve a high antimicrobial impact on pathogenic bacteria. Nonetheless, the effect of UV on extracts had never been studied on cyanobacterial extract. However, Abd-Allah et al. (2018) used UV on extracts from *Datura metel* L. and the extracts were evaluated against 3 strains of pathogenic bacteria. They found that wavelength of 250-280 nm had high optical energy and subsequently had a significant impact on the electron transitions from (π^*— n) and also (π —π^*). The extracts were even more potent and had a great antimicrobial effect (Table 2). Similarly, it was found that the antimicrobial profile was changed and actually improved when extracts were exposed to UV.

From the aforementioned facts, the exposure in case of cyanobacterial cells to UV can be mutagenic and the extent of it can vary from one strain to another (Neelam and Rai 2003) as well as being highly unpredictable. Thereby, it was recommended to use cyanobacterial extracts instead as this approach will result in enhanced antimicrobial activity without mutagenically modifying the cyanobacterial cells with UV irradiation. The enhancing effect was on extracts for isolates of diverse morphology and nature. This only emphasizes the wide potential of that approach and its applicability on diverse strain types. Hence, further exploration/exploitation of this approach on different microorganisms with interesting attributes is recommended.

Conclusion

This is the first report, to the best of knowledge, about the change in antimicrobial profile of the cyanobacterial extracts using UV treatment. The exposure was for cyanobacterial extract not to the cells themselves. This is a novel application where the extracts themselves are modified without mutagenically altering the cyanobacteria, thereby causing less hazards and giving rapid enhancement in antibacterial activity.

Abbreviation

DNA: Deoxy ribonucleic acid; KSA: Kingdom of Saudi Arabia; UV: Ultraviolet

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Ethical approval and consent to participate

Not applicable (no human materials, data, or participants were involved).

Consent of publication

Not applicable

Authors' contributions

The two authors contributed equally to the manuscript. Professor N. El Semary was responsible for conception, work design, cyanobacterial isolation, and characterization as well as the writing up. Dr. M. Aldayel was responsible for bacterial isolation, characterization and antimicrobial bioassay,

and supplying its data. They were both responsible for interpretation of data. Both authors have read and approved the manuscript.

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