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Antibacterial, Antioxidant, and Anticancer Activities of Biosynthesized Selenium Nanoparticles Using Two Indigenous Halophilic Bacteria

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Background & Aims of the Study: Selenium is an essential nutritional material used for the important functions of the human body. The issue of the production of selenium nanoparticles (SeNPs) was investigated in various fields, such as anticancer, antioxidant, and antibacterial activities.

Materials and Methods: In order to study antibacterial activity, the nutrient broth medium containing synthesized SeNPs in six different concentrations (i.e., 100, 50, 25, and 12.5 µM) was evaluated on six pathogenic bacteria. Then, the growth curves were drawn as an antibacterial assay in six pathogenic bacteria. In addition, the antioxidant effect was examined by the 2, 2-Diphenyl-1-picrylhydrazyl method. The stock solution of SeNPs was mixed with culture media in six different concentrations (i.e., 0.001, 0.01, 0.1, 1, 10, and 100 µM) and exposed on MCF-7 and HT-29 cell lines; therefore, the anticancer effects of SeNPs were assayed by the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide method.

Results: According to the obtained results, the most and least significant antibacterial effects of synthesized SeNPs were observed for *Staphylococcus aureus* (98%) and *Klebsiella pneumonia* (51.55%), respectively. The highest concentration of biosynthesized *SeNPs* can achieve a better outcome regarding the antioxidant activity. The growth of MCF-7 and HT-29 cancer cell lines were inhibited by synthesized SeNPs in a concentration of 100 µM.

Conclusion: Consequently, the results of the present study showed that SeNPs synthesized from indigenous halophilic bacteria could display anticancer, antioxidant, and antibacterial activities. This progress can assist in the treatment of different diseases.

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Background

Selenium nanoparticles (SeNPs) biosynthesis can play a predominant role in drug delivery. In addition, SeNPs have different applications in the industry, agriculture, energy, and electronics (1). The components containing the highest amount of inorganic selenium are selenocysteine, selenomethionine, and methyl-selenomethionine. Selenocysteine is an essential amino acid, and the human body could not produce it

and requires nutrients containing selenium supplement (2). It is well-known that the cells are protected from oxidative stress by glutathione peroxidase enzyme that is important for the functions of the body.

Other selenoenzymes, such as thioredoxin reductase and deiodinases, are related to removing free radicals. The selenium presence in selenoenzymes may have an important antioxidant function (3, 4). Although increasing selenium element causes toxicity in the body, its deficiency leads to different diseases associated with reproduction, thyroid and cardiovascular diseases, arthritis, muscular dystrophy, and cystic fibrosis (5).

The goal of the production of nanoparticles is the exploitation of structures and materials in size and dimension that are suitable for use, such as biological molecules. Therefore, nanoparticles can be utilized in sizes of less than 100 nm, causing their surface to increase for the target molecule, which are soluble in water. In addition, nanoparticles provide possible treatment and availability easily at the atom level (1, 6, 7). Actually, the development of nanotechnology in medicine and oncology can be a way of destroying cancer cells and assist in the treatment (8). Consequently, the importance of the production of SeNPs is related to the advances in medical and pharmaceutical sciences as well as its antioxidant and antibacterial properties (9).

The synthesis of SeNPs using biological methods usually has more advantages than other methods. The biosynthesis of SeNPs is performed using various microorganisms, such as plants, bacteria, yeasts, and fungi. The bacterial biosynthesis of SeNPs is biocompatible, eco-friendly, nontoxic, harsh-environment resistant, and easily accessible (10). The halophilic bacteria, exposed to the industrial and urban sewage (constantly), usually confront toxic metals, such as selenium, and this process can convert them to nanoparticles. *Salinicoccus iranensis* (IBRC-M 10198; Gram-positive) and *Halomonas elongate* (IBRC-M 10214; Gram-negative) are isolated from the textile industry wastewater and salt lakes in Iran and regarded as moderately halophilic and metal-resistant bacteria (11).

Currently, the antibacterial properties of SeNPs are considered among serious subjects for antibiotic-resistant bacteria worldwide (10, 12, 13). The reports have shown that SeNPs have inherent antibacterial activities leading to the removal of pathogenic bacteria (10, 12). Since SeNPs can destroy pathogen cells by several pathways (14), they could be used in the treatment of different cancers, for example, breast, colon, prostate, and lung cancers (4, 15, 16). The results of several studies have shown that reduced selenium intake is associated with the increase of cancer risk (17). The SeNPs have an effective role in the treatment of MCF7 and HT-29 cells. Baskar G et al. investigated anticancer activities of SeNPs on HT-29 and MCF7 cells (16). Depending on the size and shape of NPs, the anticancer mechanism of SeNPs varies (17).

In recent years, the problems of most people are related to the continuous and stable infection of pathogenic bacteria. Therefore, a novel method is required for the inhibition of bacteria. On the other hand, increasing the production of free radicals in the natural environment due to chemical and industrial pollution compels the researchers to develop new approaches to remove them. Consequently, treating cancer with nanoparticles is a challenging issue worldwide. With this background in mind, the present study aimed to investigate the antimicrobial, antioxidant, and anticancer effects of halophilic bacteria-originated biosynthesized SeNPs.

Materials & Methods

Chemicals

2, 2-Diphenyl-1-picrylhydrazyl (DPPH),

3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-2H-tetrazolium bromide (MTT), dimethyl sulfoxide (DMSO), Glycine, and NaCl were purchased from Sigma-Aldrich (St. Louis, MO, USA). Moreover, the RPMI-1640 medium, Trypan blue, fetal bovine serum (FBS), phosphate-buffered saline (PBS), penicillin/streptomycin solution, and trypsin-ethylenediaminetetraacetic acid solution were purchased from Gibco (Grand Island, New York).

Preparation of SeNPs

The SeNPs were produced in an optimal condition at a temperature of 37°C and 150 rpm during 72 h and 6 mM sodium of *Salinicoccus iranensis* 10198 or 8 mM sodium of *Halomonas elongata*. The synthesis of SeNPs was performed by two indigenous halophilic bacteria, namely *Halomonas elongata* (IBRC-M 10214; He10241) and *Salinicoccus iranensis* (QW6 IBRC-M 10198; *Si10198*), isolated from textile industry wastewater and Salt Lake in Qom, Iran (11). The biosynthesized SeNPs were characterized using some methods, including Ultraviolet-visible (UV-Vis) spectrophotometry, Fourier transform infrared spectroscopy (FTIR), transmission electron microscopy (TEM), Field emission scanning electron microscopy, X-ray powder diffraction (XRD), Zeta potential, and Energy-dispersive X-ray spectroscopy (EDX) (18).

Antibacterial assay of SeNPs on pathogenic bacteria

The stock solution of SeNPs with a concentration of 1,963 µM was considered for the dilution of serial preparation. In addition, the nutrient broth medium containing different SeNPs in the concentrations of 100, 50, 25, and 12.5 µM was prepared. Then, 100-µL over-night culture of Escherichia coli ATCC35218, Bacillus subtilis ATCC6051, Klebsiella pneumonia ATCC11296, Salmonella enterica ATCC9270, Pseudomonas aeruginosa ATCC25668, and Staphylococcus aureus ATCC33591 pathogenic bacteria were separately added. The pathogenic bacteria reached log phase growth; subsequently, cell suspension absorbance was measured by a spectrophotometer (PG-T80 Instrument UV-Vis Spectrophotometer, Varian, USA) at a wavelength of 600 nm. The suspension containing logarithmic culture was diluted to reach a concentration of 6.6 × 106 colony-forming unit (CFU) per milliliter of the culture medium and SeNPs.

The suspension containing 150 mL of microbe was added to each well of the microplate to obtain the cell density of 10^6 CFU/mL. Finally, it was incubated at a temperature of 37°C, and the absorbance of each well was measured at 630 nm every 2 or 4 h during 24 h using an enzyme-linked immunosorbent assay (ELISA) reader (Stat Fax 2100, UK). Subsequently, the growth curve was drawn. The control groups included positive, negative, and blank containing 150 µL of the culture media and microbe suspension as positive, culture media as negative, and nanoparticles with the culture media as blank, respectively. These control groups were formed aiming to identify exact cell density and measure the absorbance rate (19).

Antioxidant assay of SeNPs

The DPPH scavenging activity of SeNPs was investigated by the observation of the purple DPPH radical change into a yellow stable compound in the presence of an antioxidant. This reaction depends on the hydrogen donating ability of the antioxidant (20). The DPPH scavenging activity of the biogenic SeNPs was investigated based on the method described in a study by Turlo et al. with some changes (21). One milliliter of SeNPs solution (20-200 g/mL) was mixed with 1 mL of the freshly prepared DPPH solution in methanol (0.15 mM). Subsequently, the methanol (3 mL) was added, and the mixture

was incubated in dark at room temperature for 30 min. The absorbance of the mixture was recorded at 517 nm using the UV-Vis spectrophotometer.

The negative control was designed by replacing deionized water with SeNPs stock. The percentage of DPPH scavenging activity was evaluated based on the following special formula:

(Aa-Ab)/Ac ×100

where A_a is the absorbance of the sample mixed with DPPH solution; A_b is the absorbance of the sample without DPPH solution; A_c is the absorbance of the control solution. In addition, the concentration of the samples with inhibiting 50% of DPPH (half-maximum inhibitory concentration [IC_{50}]) was calculated by linear regression (22).

Anticancer assay of SeNPs

Cell culture

The MCF-7 (human breast) and HT29 (human colon) adenocarcinoma cell lines were obtained from the Pasteur Institute of Iran. The cell lines were adherently grown in 1,640 rpm medium supplemented with 10% (v/v) FBS, L-glutamine (2 mM), Penicillin G-Streptomycin (100 U/mL and 100 µg/mL) at 37ºC in a humidified incubator within 95% O_2 and 5% CO_2 (INCO 108 incubator, Memmert, Schwabach, Germany).

Preparation of concentrations of compounds and controls

Six different concentrations (0.001, 0.01, 0.1, 1, 10, and 100 µM) of the biosynthesized SeNPs and cisplatin were prepared. The stock solution of the SeNPs (1 mg/100 µL) was prepared in DMSO. Appropriate amounts of the stock (depending on molecular weight) were mixed with 1 mL of culture media to reach the concentration of 100 µM. Positive control (i.e., 1 mM hydrogen peroxide) was obtained by adding 11 µL of hydrogen peroxide (3%) to 10 mL of culture media, and negative control was the only medium. Each concentration was exposed to cell lines for 24, 48, and 72 h in at least 3 wells.

Anticancer activity

The cytotoxicity activities of compounds of SeNPs against MCF7 and HT29 cell lines were determined by MTT assay as previously described (23). Briefly, 6 × 10^3 cells/well were seeded into 96-well plates in triplicate in a medium of 200-µl volume and allowed to attach for 24 h (pre-incubation). Subsequently, 200 µL of the medium containing various concentrations of working solutions replaced the supernatant. Then, 1 mM of hydrogen peroxide solution was used as minimum viability (i.e., positive control). The medium wells were used for maximum viability (i.e., negative control). Subsequently, the plates were incubated at 37ºC for 24, 48, and 72 h.

Following the treatment, the working solution was replaced by 200 µL of a fresh medium (1/4 PBS plus 3/4 culture medium) consisting of MTT (2 mg/mL). The plates were then incubated at 37ºC in a 5% CO_2 atmosphere for 4 h. The supernatant containing MTT solution was left, and the formed violet formazan crystals were dissolved by the addition of DMSO (200 µL) and Sorensen's glycine buffer (0.1 M glycine plus 0.1 M NaCl equilibrated at pH 10.5 using 0.1 M NaOH) into each well as the stopper. Having shaken the plates for 40 min, an ELISA reader was used to determine the absorbance at 570 nm for the establishment of the cell viability.

Statistical analysis

All the data were reported as IC_{50} ± relative standard deviation (RSD=[SD/average] ×100) of at least three determinations. The growth inhibition percentage of each concentration of SeNPs was calculated using the Microsoft

Excel software (Microsoft Office 2013) by the following formula: Growth inhibition = (1 - $\frac{TestV-MinV}{MaxV-MinV}$) × 100 (1-1 equation)

The IC_{50} values (i.e., the concentrations of drug required for 50% inhibition of cell growth) were estimated by fitting the data in a sigmoidal dose-response curve by non-linear regression analysis using GraphPad Prism software (version 8.0.2) for each cell line.

Results

SeNPs synthesis and characterization

The biosynthesis of SeNPs was optimized in both bacteria, and the presence of SeNPs was confirmed by the change of the culture medium color to red. In UV-Vis spectrophotometry, a sharp absorption peak at 294 nm was observed. The spherical shaped nanoparticles were viewed in scanning electron microscopy and TEM images with a diameter of 30-100 nm. The identification of SeNPs was performed by FTIR, and the peaks were the indicators of selenium production. The highest peak diffraction in XRD analysis was related to the synthesis of SeNPs. The Zeta potential analysis showed the production of SeNPs. Then, the electron image of SeNPs by EDX confirmed elemental selenium (18).

Antibacterial effect

The antibacterial effect of biosynthesized SeNPs of halophilic bacterial species *He10214* and *Si10198* were evaluated on six different types of Gram-positive and Gram-negative pathogenic bacteria. The antibacterial effect of biosynthesized SeNPs on the growth inhibition curve in the pathogenic bacteria, namely *E. coli*, *Bacillus subtilis*, *Klebsiella pneumonia*, *Salmonella enterica*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*, was investigated in this study (figures 1 and 2).

Generally, the antibacterial effect of SeNPs synthesized by halophilic bacteria was variable based on the concentrations of SeNPs. This could be due to various mechanisms synthesizing SeNPs in these two bacterial species, namely *He10214* and *Si10198*. Based on Table 1, the observations showed that the antibacterial effect of synthesized SeNPs by *Si10198* species (in the concentration of 100 µM) on *Staphylococcus aureus* was more than those of other bacteria, and this effect was less regarding *Klebsiella pneumonia*, compared to other pathogenic bacteria. However, the SeNPs synthesized by *He10214* had the most and least antibacterial effects on *Salmonella enterica* and *Klebsiella pneumonia*, respectively.

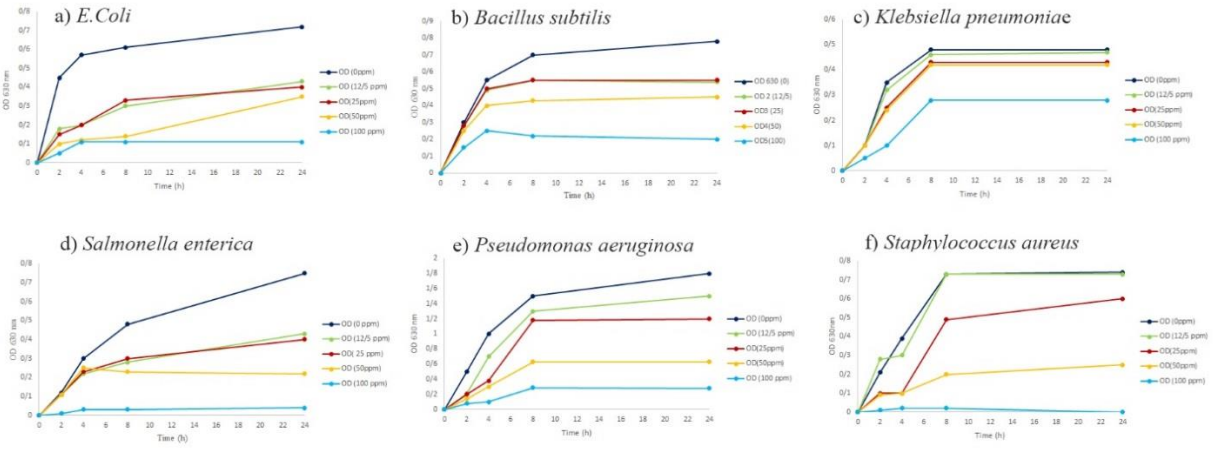

Figure 1) Growth curve evaluation of six different pathogenic bacteria by antibacterial activity of synthesized selenium nanoparticles using *Salinicoccus iranensis* strain (QW6 IBRC-M)

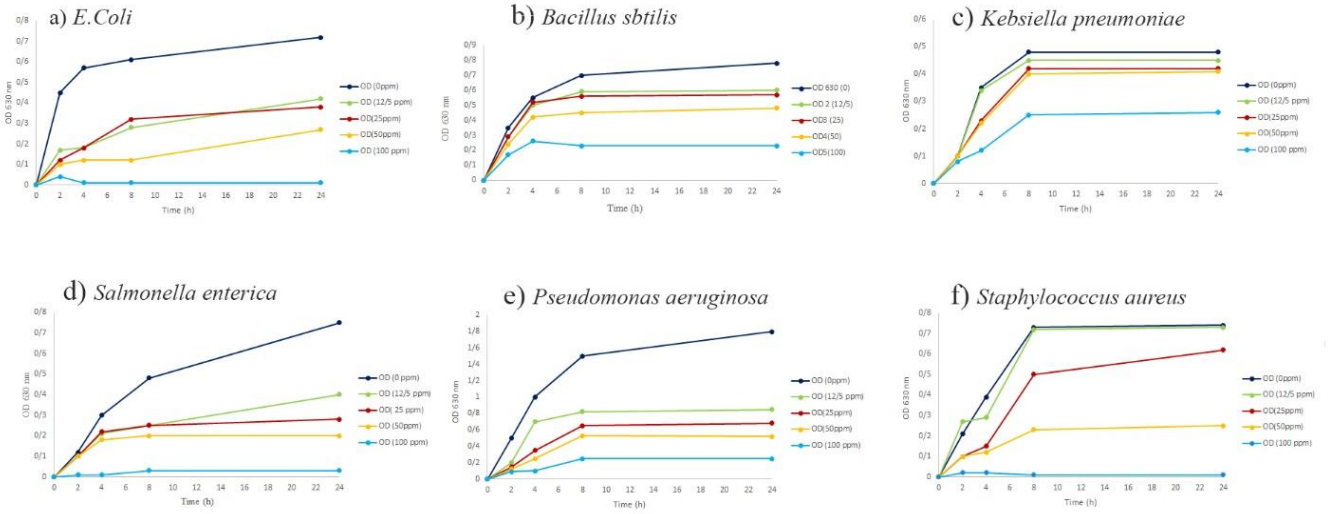

Figure 2) Growth inhibition curve evaluation of six different pathogens bacteria by antibacterial activity of synthesized selenium nanoparticles using *Halomonas elongata* (IBRC-M 10214)

Table 1) Growth inhibition of six pathogenic bacteria obtained by antibacterial effect of synthesized selenium nanoparticles by halophilic bacteria, namely *He10214* and *Si10198*

Pathogenic bacteria	Growth inhibition (%)							
	Salinicoccus iranensis (IBRC-M 10198)				*Halomonas elongata* (IBRC-M 10214)			
	12.5 (µM)	25 (µM)	50 (µM)	100 (µM)	12.5 (µM)	25 (µM)	50 (µM)	100 (µM)
***E. coli* ATCC 35218**	53.56	53.56	72.48	84.52	55.28	57	74.2	86.24
***Bacillus subtilis* ATCC 6051**	20.57	20.57	35.78	66.2	17.19	18.88	34.09	62.82
***Klebsiella pneumonia* ATCC 11296**	5.95	14.5	17.35	51.55	5.95	17.35	21.2	51.55
***Salmonella enterica* ATCC 9270**	36.82	36.82	51.4	92.71	36.82	48.97	58.69	95.14
***Pseudomonas aeruginosa* ATCC 25668**	23.36	38.35	65.01	85	46.68	64.18	70.84	85.83
***Staphylococcus aureus* ATCC 33591**	0.04	37.28	68.64	98.04	2	33.36	66.68	86.28

Antioxidant effect

The investigation on the antioxidant properties of SeNPs was performed using DPPH assay in six different concentrations (12.5, 25, 50, 100, 150, and 200 µM). The antioxidant activity of purified SeNPs in both halophilic bacterial species, *Si10198* and *He10214*, was investigated in similar conditions. The results showed that increasing the concentration of SeNPs had a significant

relationship with increasing the antioxidant effect. The antioxidant activity of SeNPs was different for both halophilic bacteria. Therefore, the most scavenging effects in the concentration of 200 µM in *Si10198* and *He10214* species were 34.6% and 33.18%, respectively (Figure 3).

Anticancer effect

The present study investigated the anticancer activity of SeNPs synthesized by *He10214* and *Si10198* species on MCF7 and HT-29 cell lines. The anticancer effect of SeNPs on MCF7 cancer cells at different times demonstrated that more than 75% of cancer cell growth was inhibited in every three replications (Figure 4).

Consequently, the highest growth change of cancer cells was defined in 72 h. According to the results of the present study, the anticancer effect of synthesized SeNPs varied depending on the organism producing them. However, the anticancer effect of synthesized SeNPs was compared in two halophilic bacteria, and the results showed that the cisplatin effect in the *He10214* bacteria was more than *Si10198*. The results of the anticancer effect of synthesized

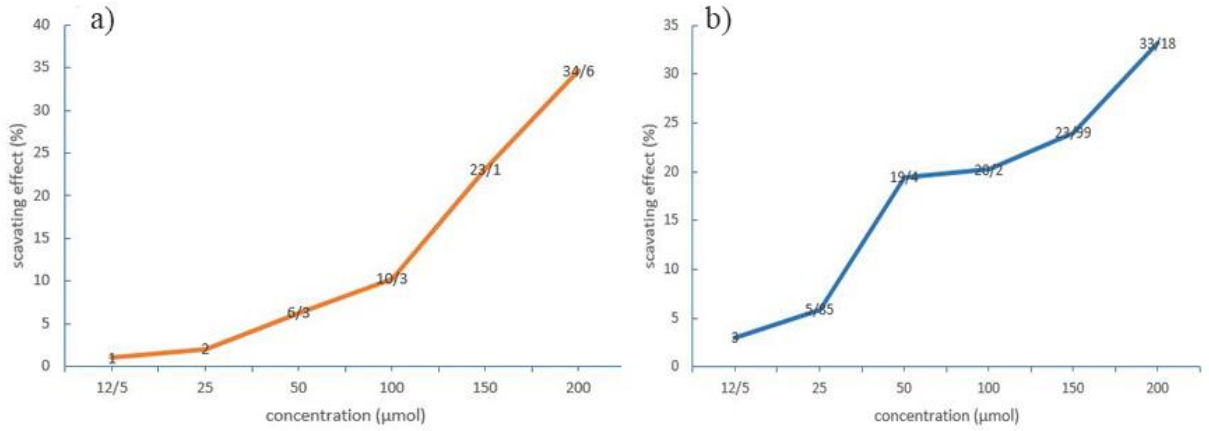

Figure 3) Evaluation of antioxidant activity of synthesized selenium nanoparticles by (a) *Salinicoccus iranensis* strain (QW6 IBRC-M 10198) and (b) *Halomonas elongata* (IBRC-M 10214)

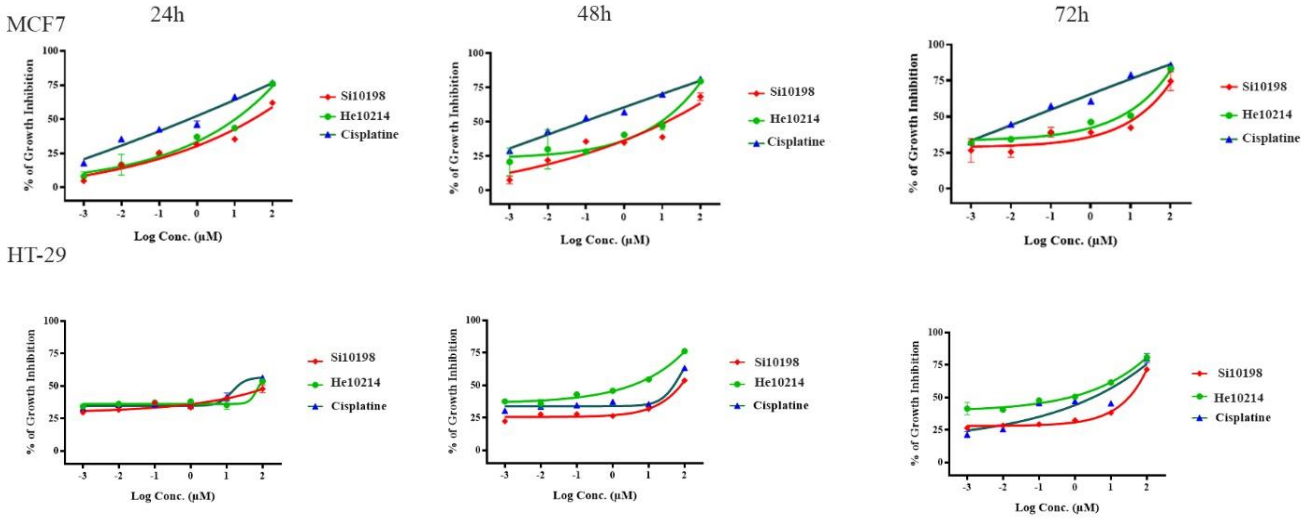

Figure 4) Comparison of anticancer activities of cisplatin and synthesized selenium nanoparticles by two halophilic bacteria (*He10214* and *Si10198*) on MCF7 and HT-29 cancer cells at different times

Table 2) Measurement of half-maximum inhibitory concentration in the investigation of anticancer effect of cisplatin and synthesized selenium nanoparticles using two *He10214* and *Si10198* bacteria

Time	IC_{50}					
	Si10198		***He10214***		**Cisplatin**	
	MCF7	**HT-29**	**MCF7**	**HT-29**	**MCF7**	**HT-29**
24 h	30.18±0.1597	>200	6.607±0.1549	>200	0.5448±0.1075	131.0±0.5669
48 h	11.30±0.2194	>200	2.865±0.3322	0.5030±0.2273	0.08194±0.07556	40.87±0.5319
72 h	3.936±0.3439	13.03±0.3493	0.7201±0.2373	0.1044±0.2379	0.03293±0.09343	1.330±0.2301

IC_{50}: Half-maximum inhibitory concentration

SeNPs on HT-29 cells related to colon cancer were successfully observed. The findings of the present study reported that the growth of HT-29 cancer cells was inhibited by the important function of SeNPs; however, a better effect was noticed on the MCF-7 cell line.

The anticancer effect of SeNPs by both halophilic bacteria at different times (i.e., 24, 48, and 72 h) was investigated in various concentrations. The growth inhibition percentage had a significant relation with increasing the concentration in such a way that the highest concentration of SeNPs (100µM) had the highest percentage of growth inhibition during 72 h. These findings were confirmed for both the MCF-7 and HT-29 cell lines. The IC_{50} determined the growth inhibition of cancer cell scale by MTT assay indicating that the concentration of SeNPs was associated with the half inhibition of cancer cell growth (Table 2).

Discussion

Nowadays, nanotechnology-based therapies are required in order to promote community health. According to multiple studies, the production of SeNPs with different methods showed important features in the functions of the body. The investigation of antibacterial, antioxidant, and anticancer effects of SeNPs demonstrated that the nanoparticles could prevent the growth of pathogenic bacteria. In addition, the MCF7 and HT-29 cancer cells in breast and colon cancers were inhibited with the anticancer effect of SeNPs. The different reactions of the body lead to the production of free radicals that selenium elements destroy with antioxidant properties.

The antibacterial effect of SeNPs biosynthesized by both bacterial species *Si10198* and *He10214* was investigated in different concentrations on pathogenic bacteria. The antibacterial effects of SeNPs depended on various concentrations of SeNPs. The growth inhibition percentage of synthesized SeNPs produced from *Si10198* and *He10214* in the six pathogenic bacteria was compared in a concentration of 100 µM. The inhibition of *E. coli*, *Pseudomonas aeruginosa*, and *Salmonella enterica* (i.e., Gram-negative bacteria) with synthesized SeNPs in the 100-µM concentration of *He10214* was more than that reported for *Si10198*.

According to the results of a study carried out by ShuBharani et al., the SeNPs biosynthesized from propolis by resazurin microtiter plate have the most antibacterial activity on *Bacillus cereus* and *Staphylococcus aureus* at a concentration of 250 µg/mL. Moreover, the moderate activity against *Salmonella enterica* was observed in the concentration of 1000 µg/mL. However, the growth of *Escherichia coli* and *Pseudomonas aeruginosa* in the high concentration of 1000 µg/mL cannot be inhibited by SeNPs (24).

The inhibition of *Bacillus subtilis* and *Staphylococcus aureus* (i.e., Gram-positive bacteria) with synthesized SeNPs in the 100-µM concentration of *Si10198* bacteria was more than that of *He10214* bacteria. Tran PA et al. have reported the antibacterial effect of

SeNPs on the growth of *Staphylococcus aureus* in four different concentrations (0, 7.8, 15.5, and 31 µg/mL) and three different times (i.e., 3, 4, and 5 h). The results have shown that SeNPs could inhibit the growth of bacteria after 3 h. In addition, the percentage of live bacteria became less after 5 h (6).

In another study, Tran PA et al. have observed that SeNPs had a strong inhibition effect on *Staphylococcus aureus*; however, there was no significant inhibition effect on *E. coli* (12). The results of the aforementioned study demonstrated that the *Klebsiella pneumonia* (i.e., a Gram-negative bacterium) inhibition with synthesized SeNPs by *He10214* bacteria in 100-µM concentration is equal to that of *Si10198* bacteria.

The antibacterial effect mechanism of SeNPs is different in various pathogenic bacteria probably related to different types of cell walls. In Gram-positive bacteria, the surface charge of the membrane is less than that of Gram-negative bacteria due to the antibacterial effect of SeNPs synthesized in various Gram-positive and Gram-negative bacteria (12). In a study conducted by Srivastava et al., it was shown that 99% growth of *P. aeruginosa*, *S. aureus*, *E. coli*, and *S. pyogenes* were inhibited in different SeNPs concentrations (25).

The inhibition effect of synthesized SeNPs by halophilic bacteria were investigated on pathogenic bacteria at the lowest concentration (12.5) in which *E. coli* species (more than 50%) was under the highest inhibition effect (Table 1). Nevertheless, the results showed that the lowest inhibition effect in the concentration of 12.5 µM was on *S. aureus* according to Table 1, and *S. aureus* had the highest inhibition in the concentration of 100 µM. Consequently, the sudden increase in the concentration of antibacterial activity of SeNPs leads to an increase in each bacterium.

The protection of body balance depends on the normal function of antioxidant components and enzymes (26). The generation of oxidative stress is related to the change of the electron transport chain function in the mitochondria inner membrane (27). Recently, the increasing biologic pollution, for example, heavy metals, certain drugs, and radiation, can remove the hemostasis of the body and enhance the production of free radicals. The development of oxidative stress leads to the destruction of body cells and biological molecules (28). The antioxidant components, such as selenium, have an important role in various functions of the body (26). The production of SeNPs with the aim of increasing the function has been taken into consideration in nanotechnology (6).

In the present study, the antioxidant activity of SeNPs was investigated by a general, rapid, and available method as DPPH (20). In a study carried out by Boroumand et al., SeNPs were produced with the chemical reduction of sodium selenite by ascorbic acid in the presence of a stabilizing agent. Boroumand et al. investigated the SeNPs antioxidant effect by the DPPH method and showed that the highest concentration of SeNPs had the highest inhibition percentage (29). Consequently, the results of the current study demonstrated that the highest concentration of SeNPs (i.e., 200 µM) had a greater effect on antioxidant activity. According to the findings of a study conducted by Shubharani et al., there is a significant relationship between the antioxidant activity of SeNPs and increasing its concentration (24). In addition, in another study performed by Vyas et al., the antioxidant activity of garlic (Allium sativum) extract was evaluated, and the inhibition effect of SeNPs in a 600-µM concentration was 67%, depending on the dose of SeNPs (30).

The anticancer activity of SeNPs on MCF7 and HT-29 cells showed that the highest anticancer effect appears after 72 h with increasing the SeNPs concentrations. For the investigation of anticancer activity, the SeNPs in six concentrations of 0.001, 0.01, 0.1, 1, 10, and 100 µM were prepared. The two types of

synthesized SeNPs by halophilic bacterial species (i.e., *He10214* and *Si10198*) and cisplatin were used to inhibit MCF7 and HT-29 cell lines.

The results of the present study showed that increasing the concentration of SeNPs caused more growth inhibition in cancer cell lines. In the investigation of the anticancer activity of SeNPs on HT-29 cells, it was observed that the effect of synthesized SeNPs by *He10214* species was higher than that of cisplatin, as a chemotherapeutic drug. This different result may be related to various behaviors of cancer cells.

According to a study carried out by Wadhwani et al., synthesized SeNPs by *Acinetobacter* sp. SW30 caused a reduction of MCF-7 viability (31). Moreover, Ranjitha and Ravishankar showed that the increase of SeNPs dose leads to an increase in its cytotoxic effect on the HT-29 cell line (32). In the current study, IC_{50} values were obtained at 3.93, 0.72, and 0.03 related to the effect of the synthesized SeNPs by halophilic bacteria of *Si10198* and *He10214* species and cisplatin, respectively. The IC_{50} on HT-29 cancer cells examined the influence of synthesized SeNPs on halophilic bacteria of *Si10198* (13.03) and *He10214* (0.1) species and cisplatin (control+) (1.33). The synthesized SeNPs function of *He10214* was better than *Si10198* that could depend on the size and dimension of SeNPs or mechanism of unknown halophilic bacteria. Therefore, this SeNPs reaction had a more considerable effect on the two MCF-7 and HT-29 type cell lines.

According to the observations, the synthesized SeNPs of *Si10198* was more effective in the MCF7 cancer cell; however, the synthesized SeNPs of *He10214* showed better effects on the HT-29 cancer cell. Probably, this difference is due to SeNPs properties and cell type; as a result, the cancer cells may be resistant to the treatment by SeNPs. The antibacterial, antioxidant, and anticancer effects of the synthesized SeNPs from *He10214* and *Si10198* species were investigated in different concentrations. The comparison of this effect demonstrated a significant relationship in a similar concentration (i.e., 100 µM).

Selenium could affect cancer cells in several different ways and inhibit reactive oxygen species (ROS) production, thiol modification, and chromatin binding and modification. In addition, the molecular processes of kinases protein signaling, activity of caspases, and p53 phosphorylation assist correct selenium function (17, 33). The anticancer effect of SeNPs depends on the intake dose and use of different selenium forms (17). The ROS generation can lead to the death of bacteria. This mechanism is a technique of the removal of pathogenic bacteria by SeNPs. Consequently, the ROS mechanism plays a key role in the important antibacterial activity and anticancer function of SeNPs (14, 34).

Conclusion

The biogenic SeNPs displayed antimicrobial effects against six pathogens bacteria, including *Escherichia coli*, *Bacillus subtilis*, *Klebsiella pneumonia*, *Salmonella enterica*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*. The highest inhibitory effects of both synthesized SeNPs were observed on the *Salmonella enterica* and *Staphylococcus aureus* species, and *Klebsiella pneumonia* showed the highest resistance to the nanoparticles. Moreover, the maximum effect of the antioxidant activity of nano-selenium was identified at the concentration of 200 µM. The present study investigated the effect of SeNPs on cancer cell lines, and the obtained results showed that their cytotoxic effects varied between different cell lines. In addition, the dose of SeNPs and duration of treatment had a very important role in their effect on the viable reduction of cancer cell lines.

Footnotes

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Conflict of Interest

The authors declare that there is no conflict of interest.

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