



## Effect of titanium dioxide nanoparticles on the polysaccharides of *Rhizobium* isolates

Hanya Khalid Al-Hayani  and Omar Abdulazeez Alhamd

Department of Life Sciences, College of Education for Pure Sciences, University of Mosul, Mosul, Iraq

Received: Apr 10, 2023/ Revised: May 4, 2023/ Accepted: May 6, 2023

(✉) Corresponding Author: [hanya.20esp45@student.uomosul.edu.iq](mailto:hanya.20esp45@student.uomosul.edu.iq)

### Abstract

This study included testing the production of *Rhizobium* isolates for external polysaccharides, and the isolates showed mixed results among them, as some isolates outperformed others in the rate of production, and also tested the effect of titanium dioxide on the production of exopolysaccharides for *Rhizobium* isolates, and the results showed that nanomaterials have an effect. There was a significant increase in the rate of production of exopolysaccharide, while the higher the concentration, the rate of production decreased significantly. The size and shape of the nanoparticles were diagnosed using SEM examination and visible spectroscopy to ensure that the nanoparticles of dioxide are within the nanoscales.

**Keywords:** *Rhizobium*, exopolysaccharides, titanium dioxide nanoparticles

### Introduction

*Rhizobium* bacteria are found in the rhizosphere, that is, around the roots of leguminous plants. Through their presence in this region, they infect plants in the root hairs. Infection often occurs through cracks on the surface of the roots of leguminous plants. As a result of the entry of *Rhizobium* bacteria into the top of the root hairs, deformities will occur. In the root hair and the formation of what is known as the infection thread (Oldroyd et al., 2011), *Rhizobium* bacteria invade neighboring cells, then the cells of the pericycle and the cortex begin to divide, leading to the formation of root nodules (Roy et al., 2020).

Root nodules are created by the secretion of the roots of leguminous plants, a mixture of chemical compounds, including Flavonoides, Isoflavonoids, and other stimuli, which are chemical signals that attract *Rhizobium*. Root nodules are divided into limited nodules and unlimited nodules depending on the plant host and the type of nodule formed in it (Andrews and Andrews, 2017).

*Rhizobium* bacteria stimulate plant growth through biological fixation of nitrogen, dissolution of phosphates and production of plant hormones (Andrews and Andrews, 2017). Capacity (Gu and Milton, 2020), that the principle of the biological

nitrogen fixation process is the reduction of atmospheric nitrogen N<sub>2</sub> to ammonia NH<sub>3</sub> in the presence of the enzyme Nitrogenase (Yang et al., 2018).

*Rhizobium* bacteria produce three different types of polysaccharides. Many recent studies indicated that polysaccharides are present on the surface of the bacterial cell, which are important in establishing the symbiotic relationship between leguminous plants and *Rhizobium* bacteria. These sugars are:

Capsular polysaccharides (KP)

Lipopolysaccharides (LPS)

Exopolysaccharides (EPS)

These sugars may be present on the surface of the cell, or they may be present in the space around the plasma membrane, and some of them are secreted into culture media (Lepek and D'Antuono, 2005).

The role of exopolysaccharide in the formation of the symbiotic relationship between bacteria and the host plant is important and necessary, as its mechanical role is to protect bacterial cells from harsh or adverse environmental conditions

(Dieltjens et al., 2020), and EPS has an important role in the first events Infection of the root nodes of the host plant represented by the adhesion of *Rhizobium* bacteria to the roots of the host plant, as well as its important role in the formation of the infection thread, after which the bacteria are liberated from the infection thread and the development of the formation of Bacteroids and the suppression of the host plant's defense responses (Frayssse et al., 2003), and they are also It protects the microbial cell (*Rhizobium*) against antimicrobial compounds and heavy metals (Mohite et al., 2017).

The EPS produced by the bacteria helps fix atmospheric nitrogen by lowering the high oxygen pressure (Kumari et al., 2009), and it also works to change the form of *Rhizobium* bacteria from the stick form to the form of nitrogen-fixing known as Bacteroids within the root nodules of host plants (Frayssse et al., 2003).

Several studies have shown that NPs are toxic to many microorganisms in groundwater and soil, ultimately affecting the entire food chain (Colvin, 2003).

Studies have also shown that silver nanoparticles are toxic to many different bacterial species (Dorobantu et al., 2015), and NPs are now widely used in many fields of science, but silver is the most widely used in this field, as NPs generally affect In the legume-*Rhizobium* symbiosis by resolving the number and size of the nodules are reduced gene of expression and with increasing concentration of NPs the total length of treated plants decreased (Abd-Alla et al., 2016), titanium is a shiny silvery-white metal, with low density and high strength, stronger than steel. But lighter than it is, titanium resists corrosion by forming a thin, stable, dense, and adhesive layer of Oxide when exposed to air, therefore, oxygen is required in the surrounding medium (Graciani et al., 2009).

TiO<sub>2</sub> particles are insoluble in water, hydrochloric acid, or organic solvents, meaning that they are very stable and only slightly decompose according to conditions, including low pH (Gomaa et al., 2015).

Titanium dioxide nanoparticles are synthesized by the electrochemical oxidation process, by anodizing titanium sheets in an ethylene glycol solution (Heidari et al., 2019), and the toxicity of titanium nanoparticles towards cancer cell lines is increasing, as a result of the deposition of platinum nanoparticles on them (Baqer et al., 2020), and many studies have shown that there are four different ways of exposure to TiO<sub>2</sub>NPs in humans and animals, which are ingestion, pulmonary absorption (by inhalation), and through skin or injection, and among these methods is the absorption of TiO<sub>2</sub>NPs and it is distributed by blood Or the lymphatic system and reach different parts of the body, including the heart, liver, kidneys, pancreas, bones, or nervous system, where it is preserved and can become toxic (Shakeel et al., 2016).

## Methods and material

### Prohibited culture media

#### Yeast extract manitolmedium (YEM)

This medium is prepared by dissolving (a) gm of yeast extract, 10 gm of manitol, 0.20 gm of MgSO<sub>4</sub>×7H<sub>2</sub>O, 0.50 gm of KH<sub>2</sub>PO<sub>4</sub>, 0.10 gm of NaCl, 12 gm of agar in a liter of distilled water (Dubey and Maheshwori, 2002).

#### Trypton yeast extract medium (TY)

(TY) medium was prepared by dissolving 3 gm of yeast extract, 5 gm of Trypton, and 0.12 gm of CaCl<sub>2</sub>.2H<sub>2</sub>O in a liter of distilled water, and adjusting the pH at 7.2, then autoclaving (Khanuja and Kumar, 1988).

### Exopolysaccharide production test for *Rhizobia* isolates

#### Preparation of vaccines for rhizobial isolates

Vaccines for rhizobial isolates were prepared by transferring part of the culture of isolates growing on TY agar medium to a conical flask with a capacity of 100 ml containing 10 ml of liquid YEM medium, and the flasks were incubated in the New Brunswick scientific rocking incubator at a rate of 150 revolutions/min. At a temperature of 28±2°C and for a period of 48 (Ghosh and Basu, 2001).

#### Reparation of the culture medium for the production of exopolysaccharides

The liquid YEM nutritional medium was prepared for the development of *Rhizobium* isolates and the production of exopolysaccharides, and the pH was set at 6.8. The flasks were sealed with a cotton stopper, then sterilized by autoclave at 1 atmosphere and at a temperature of 121°C for a period of 20 minutes, after which the flasks were left to cool down, then inoculated with 2% rhizobial inoculum, then the flasks were placed in the rocking incubator at a temperature of 28 ±2°C for 48 hours and a rate of 150 cycles/minute (Ghosh and Basu, 2001).

#### Extract and estimate the mass of EPS

The EPS mass is extracted and estimated by the following method, where the glass flasks are withdrawn from the rocking incubator containing the liquid bacterial culture after incubating it for 48 hours, and the liquid of the bacterial culture is centrifuged using a center fuge centrifuge at a rate of 6000 revolution/minute for some time, 20 minutes and the clearing is left aside, while the sediment formed at the bottom of the centrifuge tube is discarded, and then 5 ml of the clearing resulting from the centrifugation process free of bacterial cells is withdrawn and 10 ml of acetone solution is added for each 5 ml of the filter (Navarini et al., 1997), The

mixture was mixed well, and then a second centrifugation process was carried out at a rate of 9000 revolutions/minute for 30 minutes, at this stage the liquid was discarded and the precipitate represented by the exo polysaccharides was collected in dishes of known weight, then the dishes were placed in an electric oven at a temperature 80°C for 24 hours, and after drying the plates, the EPS mass is calculated by the difference of the two masses (Duta et al., 2006).

### The effect of nanoparticles on the production of exopolysaccharide of *Rhizobia* isolates

In the beginning, nano solutions were prepared from ready-made nanopowder in three concentrations 250, 500, and 750 mg / l, where they were dissolved in sterile distilled water, sterilized in a water bath at 60°C for half an hour, and placed in an ultrasonic device for 10 minutes. This device breaks down the nanoparticles so that they do not remain aggregated and obtain a clear nano-solution without precipitation (Fan et al., 2014).

The liquid YEM medium was prepared in test tubes, sterilized and left to cool, then 0.1 ml of the prepared nanoparticles solution was added with three concentrations of the three nanoparticles sterilized in the water bath, and then 0.1 ml of the liquid rhizobial culture was added and placed in the shaking incubator at a temperature of  $28 \pm 2^\circ\text{C}$  for a period of 48 hours and at a rate of 150 revolutions/minute to develop rhizobia isolates with nanoparticles to see their ability to produce exopolysaccharides (Oves et al., 2014). To find out the ability of the isolates to produce sugar, the usual method for extracting and estimating EPS is used as shown above.

### Examination of nanoparticles using a scanning electron microscope (SEM)

To study the shape and surface topography of the nanoparticles, the nanopowders were sent for SEM examination, and the device used was Inspect F50-FE=SEM in Iran.

### Examination of the visible-ultraviolet spectrum

The metallic nanoparticles were identified using an ultraviolet spectroscopy device (Alfuraydi et al., 2019), as a beam with a wavelength ranging between 180-1100 nm was passed through the nano-solution, and the sample placed in the test tube usually made of quartz for analysis was absorbed. Ultraviolet light (Islam et al., 2017), where 2 ml of the nanoparticle solution was placed in a quartz tube with a thickness of 1 cm, and the reading was recorded using a UV device that operates within a range between 200-800 nanometers.

## Results and discussion

### Production of exopolysaccharide by studied local rhizobial isolates

The aforementioned method of work was adopted, and after two days of incubation in the shaking incubator, the EPS production rate was estimated as shown in Table 1.

Depending on the results of the Table 1 in which there is a discrepancy or difference in the rate of exo polysaccharide production for each isolate from the others, as these results agreed with the results of the researcher (Navarini et al., 1997) in the variation in the production of external polysaccharide isolates, and the researcher relied on The method described in paragraph (10-3) of the third chapter was also based on visual observations, that is, using the naked eye, where he chose the isolate R.hedysari HCNTI as the best isolate that produces external polysaccharides, after cultivating the samples on the YEM liquid food medium and incubating for 48 hours. The hour and the researcher studied (Primo et al., 2019) The productivity of the exopolysaccharide of the bacterium *Enisifer meliloti* for isolates isolated from the environment and other laboratory isolates and studied the conditions of bacterial inoculation of the host plant, as it was found that the cell surface molecules, including the exo polysaccharide, have an important and distinctive role in the symbiotic relationship between the *Rhizobium* bacterium and the host plant And it was shown that the gene expR, which gives mucus texture to the growth of *Rhizobium* bacteria, had an effective effect on the production of exopolysaccharides.

### Effect of nanoparticles on the production of exopolysaccharide of *Rhizobia* isolates

For silver nanoparticles, and according to the tables installed in the fourth chapter, the results converged with the results of the researcher (Kaur et al., 2021), and for titanium dioxide, the results of our study converged with the results of the researcher's study (Oves et al., 2014).

Through the results that appeared to us, it was found that the production of EPS under the influence of nanomaterials of all kinds had decreased in the high concentration of 750 mg / L, because the higher the concentration of the nanomaterials, the greater the inhibition of bacterial growth, and that external polysaccharides are present on the surface of the bacterial cell, or in the space between the plasma membrane (Lepek and D'Antuono, 2005). The nanomaterials, as a result of their disintegration and liberation of their ions, will work to dissolve sugars present on the surface of the bacterial cell, which reduces the sugars secreted from rhizobial isolates (Sivasankar et al., 2018), and the results of the study (Wu et al., 2010) proved that nanomaterials can enhance Formation of exopolysaccharides as indicated in the Table 2.

**Table 1. Exogenous polysaccharide production of the studied local *Rhizobium* isolates**

<i>Rhizobium</i> isolates		Exogenous polysaccharide production		
		Tare weight	Plate weight after extraction	Average weight
HA1 <i>Ensifer fredii</i> bv. <i>Fredii</i>		6.4923	6.4947	6.4935
		.001150	.000580	.001520
HA2 <i>Ensifer fredii</i> bv. <i>Fredii</i>		7.1503	7.1540	7.1522
		.000580	.001000	.002140
HA3 <i>Ensifer fredii</i> bv. <i>Fredii</i>		7.1523	7.1557	7.1540
		.001150	.000580	.002000
HA4 <i>Ensifer fredii</i> bv. <i>Fredii</i>		7.1563	7.1597	7.1580
		.001150	.000580	.002000
HA5 <i>Ensifer fredii</i> bv. <i>Fredii</i>		6.1467	6.1493	6.1480
		.000580	.001150	.001670
HA6 <i>Ensifer fredii</i> bv. <i>Fredii</i>		6.4957	6.4993	6.4975
		.001150	.000580	.002170
HA7 <i>Ensifer meliloti</i>		7.1553	7.1587	7.1570
		.000580	.000580	.001900
HA8 <i>Ensifer meliloti</i>		6.4913	6.4950	6.4932
		.001150	.001000	.002230
HA9 <i>Ensifer meliloti</i>		6.1577	6.1623	6.1600
		.001150	.000580	.002680
HA10 <i>R. leguminosarum</i> bv. <i>Phaseoli</i>		6.1413	6.1453	6.1433
		.001150	.001150	.002420
HA11 <i>R. leguminosarum</i> bv. <i>Phaseoli</i>		7.1203	7.1227	7.1215
		.000580	.000580	.001380
HA12 <i>R. leguminosarum</i> bv. <i>Phaseoli</i>		7.1333	7.1353	7.1343
		.000580	.000580	.001210
HA13 <i>R. leguminosarum</i> bv. <i>Phaseoli</i>		7.1507	7.1563	7.1535
		.001150	.002520	.003560

**Table 2. Effect of TiO<sub>2</sub>NPs on the production of exopolysaccharides of *Rhizobium* bacteria isolates**

<i>Rhizobium</i> bacteria isolates	Effect of TiO <sub>2</sub> NPs on the production of exopolysaccharides	
	Tare weight	Plate weight after extraction
<b>250 mg/L</b>		
HA1 <i>Ensifer fredii</i> bv. <i>Fredii</i>	7.1513	7.1543
	.001150	.001150
HA5 <i>Ensifer fredii</i> bv. <i>Fredii</i>	6.1907	6.1937
	.001150	.001150
<i>Rhizobium leguminosarum</i> bv. <i>viciae</i> HA19	6.1420	6.1453
	.001730	.000580
Average	6.4947	6.4978
	.492950	.492860
<b>500 mg/L</b>		
<i>R. leguminosarum</i> bv. <i>phaseoli</i> HA10	6.4940	6.4977
	.001730	.001150
<i>Ensifer meliloti</i> HA18	7.1307	7.1363
	.001150	.001150
<i>Rhizobium leguminosarum</i> bv. <i>viciae</i> HA24	7.1507	6.8190
	.000580	.583700
Average	6.9251	6.8177
	.323450	.402070
<b>750 mg/L</b>		
HA8 <i>Ensifer meliloti</i>	6.1507	6.1543
	.000580	.001150
Average	6.1507	6.1543
	.000580	.001150



### Examination of nanoparticles using a scanning electron microscope

The shape and size of the nanoparticles were diagnosed using a scanning electron microscope, where the titanium dioxide nanoparticles appeared in almost spherical shapes with the

presence of agglomerates and sizes ranging between (36, 72) nm, The titanium dioxide nanoparticles appeared in the scanning electron microscope (SEM) as shown. These results were consistent with the results of the researcher (Saranya et al., 2018), where the particles appeared in irregular spherical shapes.

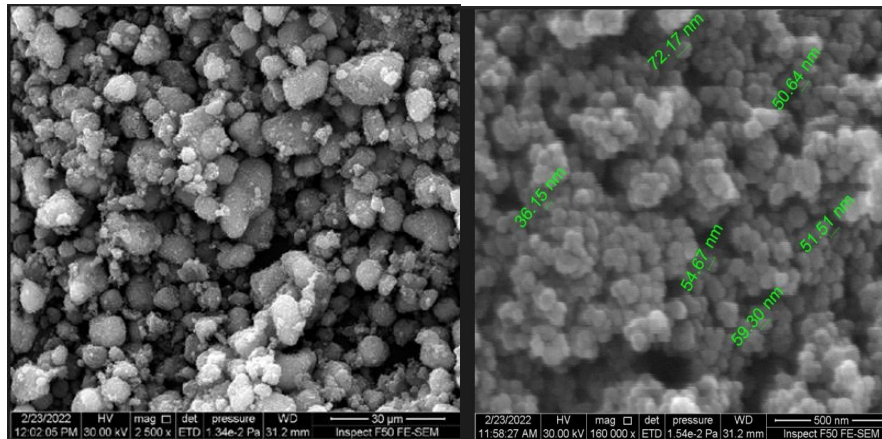


Fig. 1 shows the size of titanium dioxide nanoparticles using a scanning electron microscope

### Examination of the visible-ultraviolet spectrum

The results of the ultraviolet and visible spectrum examination showed the appearance of different absorption peaks for each nano-solution. The results of the examination of the nano-titanium dioxide solution showed the highest absorption peak at the wavelength of 970 nm, as shown in Fig. 1. The result of the nano-titanium dioxide solution converged with Findings of the researcher (Durairaj et al., 2014).

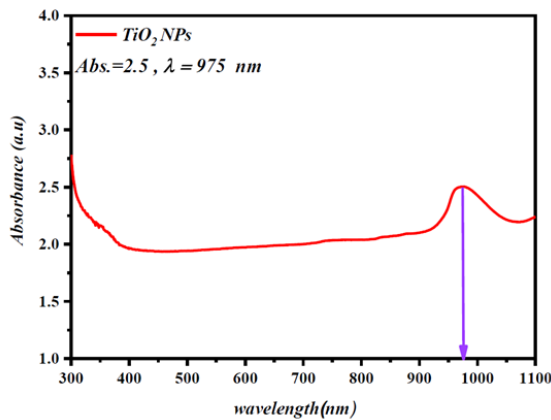


Fig. 2 Absorption values of nanoparticles using a UV-vis spectrophotometer

### Conclusions

The possibility of obtaining *Rhizobium* isolates with abundant production of exopolysaccharides, which can be used in various industrial, agricultural and medical applications. The

effect of nanoparticles in reducing or preventing the formation of exo polysaccharides in isolates of *Rhizobium* bacteria.

### Conflict of Interest

The author declares that there are no conflicts of interest.

### Consent for publication

The author declares that the work has consent for publication.

### Funding support

The author declares that they have no funding support for this study.

### References

- Abd-Alla, M. H., Nafady, N. A., & Khalaf, D. M. (2016). Assessment of silver nanoparticles contamination on faba bean-Rhizobium leguminosarum bv. viciae-Glomus aggregatum symbiosis: Implications for induction of autophagy process in root nodule. *Agriculture, Ecosystems & Environment*, 218, 163-177.
- Alfuraydi, Akram A; Al-Ansari, M., Alkubaisi, N., Gopinath, K., Karthika, V., Arumugam, A., & Govindarajan, M. (2019). Facile and cost-effective Ag nanoparticles fabricated by liliun lancifolium leaf extract: Antibacterial and antibiofilm potential. *Journal of Cluster Science*, 30, 1081-1089.
- Andrews, M., & Andrews, M. E. (2017). Specificity in legume-rhizobia symbioses. *International journal of molecular sciences*, 18(4), 705.
- Baqer, S. R., Alsammaraie, A. M. A., Alias, M., Al-Halbosiy, M. M., & Sadiq, A. S. (2020). In Vitro Cytotoxicity Study of Pt Nanoparticles Decorated TiO<sub>2</sub> Nanotube Array. *cancer*, 17(4), 1169-1176.
- Colvin, V. (2003). Responsible nanotechnology: Looking beyond the good news. *www.eurekalert.org*.
- Dieltjens, L., Appermans, K., Lissens, M., Lories, B., Kim, W., Van der Eycken, E. V., ... & Steenackers, H. P. (2020). Inhibiting bacterial cooperation is an evolutionarily robust anti-biofilm strategy. *Nature Communications*, 11(1), 107.

- Dorobantu, L. S., Fallone, C., Noble, A. J., Veinot, J., Ma, G., Goss, G. G., & Burrell, R. E. (2015). Toxicity of silver nanoparticles against bacteria, yeast, and algae. *Journal of nanoparticle research*, 17, 1-13.
- Dubey, R. C., & DK, M. (2002). A Textbook of Practical Microbiology. S. Chand & Co. Ltd, Ram Nagar, Newdelhi, 1, 2-192.
- Durairaj, B., Xavier, T., & Muthu, S. (2014). Fungal-generated titanium dioxide nanoparticles for UV Protective and bacterial-resistant fabrication. *International Journal of Engineering Science and Technology*, 6(9), 621.
- Duta F. P., Pessôa de França, F., & de Almeida Lopes, L. M. (2006). Optimization of culture conditions for exopolysaccharides production in *Rhizobium* sp. using the response surface method. *Electronic Journal of Biotechnology*, 9(4), 0-0.
- Fan, R., Huang, Y. C., Grusak, M. A., Huang, C. P., & Sherrier, D. J. (2014). Effects of nano-TiO<sub>2</sub> on the agronomically-relevant *Rhizobium*-legume symbiosis. *Science of the Total Environment*, 466, 503-512.
- Frayse, N., Couderc, F., & Poinso, V. (2003). Surface polysaccharide involvement in establishing the rhizobium-legume symbiosis. *European Journal of Biochemistry*, 270(7), 1365-1380.
- Ghosh, A. C., & Basu, P. S. (2001). Extracellular polysaccharide production by *Azorhizobium caulinodans* from stem nodules of leguminous emergent hydrophyte *Aeschynomene aspera*.
- Gomaa, I. E., Abdel Gaber, S. A., Bhatt, S., Liehr, T., Gleis, M., El-Tayeb, T. A., & Abdel-Kader, M. H. (2015). In vitro cytotoxicity and genotoxicity studies of gold nanoparticles-mediated photo-thermal therapy versus 5-fluorouracil. *Journal of Nanoparticle Research*, 17, 1-11.
- Graciani, J., Hamad, S., & Sanz, J. F. (2009). Changing the physical and chemical properties of titanium oxynitrides TiN 1-x O x by changing the composition. *Physical Review B*, 80(18), 184112.
- Gu, W., & Milton, R. D. (2020). Natural and engineered electron transfer of nitrogenase. *Chemistry*, 2(2), 322-346.
- Heidari Khoei, M., Khoei, S., & Lotfi, M. (2019). Synthesis of titanium dioxide nanotubes with liposomal covers for carrying and extended release of 5-FU as anticancer drug in the treatment of HeLa cells. *Analytical biochemistry*, 572, 16-24.
- Islam, Z. A., Mondal, S., & Islam, M. (2017). Applications, synthesis and characterization of gold nano particles (Doctoral dissertation, BRAC Univeristy).
- Kaur, H., Kalia, A., & Sharma, S. P. (2021). Multi-Wall Carbon Nanotubes, Metal Oxide and Hydroxy-Apatite Nanoparticles Enhanced Plant Growth Promoting Capabilities of Root Endosymbionts of Cowpea (*Vigna unguiculata* (L.) Walp.). *Journal of Nanoscience and Nanotechnology*, 21(6), 3634-3649.
- Khanuja, S. S., & Kumar, S. (1988). Isolation of phages of *Rhizobium meliloti* AK631. *Indian journal of experimental biology*, 26(9), 665-667.
- Kumari, B. S., Ram, M. R., & Mallaiiah, K. V. (2009). Studies on exopolysaccharide and indole acetic acid production by *Rhizobium* strains from Indigofera. *African Journal of Microbiology Research*, 3(1), 10-14.
- Lepek, V. C., & D'Antuono, A. L. (2005). Bacterial surface polysaccharides and their role in the rhizobia-legume association. *Lotus Newslett*, 35, 93-105.
- Mohite, B. V., Koli, S. H., Narkhede, C. P., Patil, S. N., & Patil, S. V. (2017). Prospective of microbial exopolysaccharide for heavy metal exclusion. *Applied biochemistry and biotechnology*, 183, 582-600.
- Navarini, L., Stredansky, M., Matulova, M., & Bertocchi, C. (1997). Production and characterization of an exopolysaccharide from *Rhizobium hedysari* HCNT 1. *Biotechnology letters*, 19(12), 1231-1234.
- Oldroyd, G. E., Murray, J. D., Poole, P. S., & Downie, J. A. (2011). The rules of engagement in the legume-rhizobial symbiosis. *Annual review of genetics*, 45, 119-144.
- Oves, M., Khan, M. S., Zaidi, A., Ahmed, A. S., & Azam, A. (2014). Production of plant-growth promoting substances by nodule forming symbiotic bacterium *Rhizobium* sp. Os1 is influenced by CuO, ZnO and Fe<sup>3+</sup> nanoparticles. *The IIOAB Journal*, 5(4), 1.
- Primo, E. D., Cossovic, S., Nievas, F., Bogino, P., Humm, E. A., Hirsch, A. M., & Giordano, W. (2020). Exopolysaccharide production in *Ensifer meliloti* laboratory and native strains and their effects on alfalfa inoculation. *Archives of microbiology*, 202, 391-398.
- Roy, S., Liu, W., Nandety, R. S., Crook, A., Mysore, K. S., Pislariu, C. I. & Udvardi, M. K. (2020). Celebrating 20 years of genetic discoveries in legume nodulation and symbiotic nitrogen fixation. *The Plant Cell*, 32(1), 15-41.
- Saranya, K. S., Vellora Thekkai Padil, V., Senan, C., Pilankatta, R., Saranya, K., George, B., ... & Āernik, M. (2018). Green synthesis of high temperature stable anatase titanium dioxide nanoparticles using gum kondagogu: characterization and solar driven photocatalytic degradation of organic dye. *Nanomaterials*, 8(12), 1002.
- Shakeel, M., Jabeen, F., Shabbir, S., Asghar, M. S., Khan, M. S., & Chaudhry, A. S. (2016). Toxicity of nano-titanium dioxide (TiO<sub>2</sub>-NP) through various routes of exposure: a review. *Biological trace element research*, 172, 1-36.
- Sivasankar, P., Seede, P., Poongodi, S., Sivakumar, M., Murugan, T., Sivakumar, L. & Balasubramanian, T. (2018). Characterization, antimicrobial and antioxidant properties of exopolysaccharide mediated silver nanoparticles synthesized by *Streptomyces violaceus* MM72. *Carbohydrate polymers*, 181, 752-759.
- Wu, B., Huang, R., Sahu, M., Feng, X., Biswas, P., & Tang, Y. J. (2010). Bacterial responses to Cu-doped TiO<sub>2</sub> nanoparticles. *Science of the total environment*, 408(7), 1755-1758.
- Yang, J., Xie, Z., Tian, Z., Dixon, R. and W. Y. (2018). Polyprotein strategy for stoichiometric assembly of nitrogen fixation components in synthetic biology. *pans.*, 115(36): 8509-5817.

#### How to cite this article

Al-Hayani, H. K. and Alhamd, O. A. (2023). Effect of titanium dioxide nanoparticles on the polysaccharides of *Rhizobium* isolates.. *Science Archives*, Vol. 4(2), 106-111. <https://doi.org/10.47587/SA.2023.4207>

This work is licensed under a [Creative Commons Attribution 4.0 International License](https://creativecommons.org/licenses/by/4.0/)



**Publisher's Note:** MD International Publishing stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.