

Biosynthesis of Iron Oxide Nanoparticles and Combination with Glyphosate Herbicide and Effect Against Cogon Grass Control

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Abstract

For enhancing herbicide efficacy a total of 42 soil samples, five species of bacteria that grew the most were selected for regarding other isolates: (*Enterobacter cloacae*, *Serratia fonticola*, *Aeromonas hydrophila*, *Pantoea* spp., *Pseudomonas mendocina*). Extracellular extracts of these bacteria were made and then used to manufacture iron oxide nanoparticles biologically. It was observed after measuring the average diameter by AFM (Atomic Force Microscopy) for each NPs prepared were 65.14, 29.21, 63.87, 57.89 and 36.59 nm, respectively. The synthesis conditions were a pH of 7 and 50°C. Based on the AFM values, *Serratia fonticola* was selected as having the smallest average diameter of 29.21 nm to complete the rest of the techniques (UV-VIS, AFM, FTIR and FE-SEM). The wavelength of Fe₂O₃NPs by using UV-VIS is 359 nm, Image FE-SEM displays spherical Fe₂O₃NPs in Nano-cluster form and the average volume is 29.21 nm by AFM. Utilizing the Randomized Complete Block Design methodology with 6 treatments, two factors: Glyphosate herbicide at the recommended dose and three concentrations of the prepared (Fe₂O₃NPs) from *Serratia fonticola* were 0.5, 1 and 1.5 g/ml. Halfa plant was controlled using these treatments, and after 30 days, the efficiency of the treatments was measured on a set of characteristics. It was found the lowest number of plants and the highest percentage of control was in the treatment 5 glyphosate and Fe₂O₃ NPs con. (1.5g/ml) was 1.26, 5.33 and 90%, respectively. The study effectively shows the potential of employing Fe₂O₃NPs generated from *S. fonticola* to improve the efficacy of Glyphosate for Cogon management.

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Introduction

The soil is a vast reservoir of microbial organisms, containing a diverse array of bacterial species that are essential for maintaining ecological balance and the overall health of the surrounding environment (Christian *et al.*, 2008). Conventional approaches utilizing culture-dependent techniques have been utilized to evaluate the microbial makeup of soil. However, these approaches only allow access to a small fraction of soil bacteria, approximately 0.1-1%. As a result, the vast phylogenetic diversity of soil

bacteria remains largely unexplored (Zhang and Xu, 2008).

Nanotechnology involves the creation of materials at the Nanoscale and the precise manipulation of particle shape and size throughout their development. Nanometer-scale materials have at least one dimension that is less than 100 nm and display a wide variety of geometric shapes, such as plates, sheets, tubes, wires, and particles (Bogunia-Kubik and Sugisaka, 2002; Goodsell, 2004). Nanoparticles play a vital role in various technologies, including healthcare, cosmetics, agriculture, and food sciences (Kingsley, 2013). Green

nanotechnology employs biological mechanisms to synthesize nanomaterials, hence reducing the generation of toxic compounds. Conversely, green nanotechnology requires significantly less energy input compared to other technologies. Additionally, the synthesis process generates few hazardous compounds, resulting in a high level of environmental compatibility. Consequently, the green nanomaterials that are manufactured have the potential for extensive utilization across diverse industries (Bartolucci *et al.*, 2020).

Various microorganisms, including bacteria, fungi, yeasts, and microalgae, have been reported to produce MtNPs either internally or externally. These bacteria possess the capability to synthesize organic substances internally and export them outside of their cells (Mandal *et al.*, 2006). Among the different methods for environmentally friendly production of Mt NPs, the ones that carry out extracellular synthesis are particularly intriguing. This is because the external placement of the material removes the requirement for expensive and intricate measures in the later stages to retrieve nanoparticles from within the cells (Singh *et al.*, 2016).

The utilization of ecologically friendly raw materials, such as biological extracts derived from bacteria, for the production of iron oxide nanoparticles offers numerous economic and compatible benefits (Christian *et al.*, 2008; Al-Azawi *et al.*, 2019). Nano-scale iron particles refer to iron particles that have a size smaller than one micron. Due to their extensive surface area, they exhibit high reactivity. When oxygen and water are present, they rapidly undergo oxidation, resulting in the generation of free iron ions.

Magnetic iron oxide nanoparticles are the preferred option for biological and medical specialized applications because of their biocompatibility, super paramagnetic properties, and chemical stability (Mahdavi *et al.*, 2013).

First described by (Gavini *et al.*, 1979) *Serratia fonticola*, a constituent of the Enterobacteriaceae family, is prevalent in many habitats such as drinking water, soil, and

sewage. Nanotechnology has the potential to improve agricultural operations through the use of nanoparticle-based fertilizers and by promoting plant growth, hence enhancing soil quality and the overall quality of agricultural products. Moreover, the use of fertilizers and insecticides that rely on nanoparticle-based carriers and chemicals is reduced without affecting productivity (Duhan *et al.*, 2017).

The plant known as Cogon grass (*Imperata cylindrica* L.) belongs to the Poaceae family. It is a warm-season weed. Its origin is South Asia. It is widespread in the countries of the Middle East and West Africa. It is considered one of the most dangerous weeds in orchards, non-agricultural lands, and roadsides (Garritty *et al.*, 1997; Terry *et al.*, 1997). It also spreads along irrigation canals and waterways (MacFaddin, 2000). It is one of the difficult-to-control weeds and is characterized by its high competitive ability with plants that grow with it by forming a network of rhizomes that spread under the surface of the soil. On the other hand, the plant is characterized by its rapid growth and the vegetation structure is thick and it's high at 120-175 cm and can grow again when exposed to unnatural conditions (Omezine and Fethia, 2009). The Halfa grass in Iraq is one of the dangerous wilds that spreads in orchards, especially newly established ones (Al-Jubouri, 1978). Glyphosate herbicide has been proven to be very effective in reducing its risk compared to other systemic pesticides (Chikoye *et al.*, 2002; Teuton *et al.*, 2005). The efficiency of herbicide use depends mainly on the amount absorbed and transferred to active growth sites in plants (Al-Chalabi and Maad, 2004).

Nano pesticide formulations can enhance the solubility of water, boost bioavailability, and provide protection against environmental degradation for agrochemicals. This has the potential to dramatically change the use of diseases, weeds, and pests in crops. However, the properties of nanomaterials are also approaching the limits of potential loss of cellular and genetic material (Yadav *et al.*, 2020).

The main objectives of this study were twofold: first, to investigate the bacterial species

commonly found in the studied soil and second, to investigate the biological properties of iron oxide nanoparticles (Fe_2O_3 NPs) administered extracellular extract from *Serratia fonticola*. Moreover, synergistic effects of iron oxide nanoparticles (Fe_2O_3 NPs) and glyphosate herbicides in the management of cogon weed (halfa).

Material and Methods

In this study, forty-two soil samples were collected from eight different fields in Baghdad which were four regions each for Al-Karkh and Al-Rusafa. The isolation and identification of group bacteria propagated in soil samples was based on biochemical assays and the Vitek2 system used to identify each bacteria at different times from February to June 2022.

Identification of bacterial isolate by Vitek-2 compact System

The VITEK® 2 Compact system is an advanced technology used in laboratories for rapid and accurate detection of microorganisms. It is designed to improve laboratory efficiency and increase productivity, due to its extensive and complex specifications (Teuton *et al.*, 2005).

Extracellular processes

Bacteria were cultured in nutrient broth for 48 hours at 28°C, then centrifuged, and the bacterial supernatant produced an extracellular extract seen as a faint yellow precipitate through the broth (Moore *et al.*, 2010).

Production of Fe_2O_3 iron oxide nanoparticles by the extracellular extract of *Serratia fonticola*

The present study focuses on the synthesis of iron oxide nanoparticles from a biotechnical point of view, using ferric sulfate- $\text{Fe}_2(\text{SO}_4)_3$ (Indian) as a precursor with some modifications (Yaaqob, 2022), for S Synthesis of Fe_2O_3 nanoparticles. The extracellular product solution obtained from *S. fonticola* was diluted in 50 ml to prepare a solution containing 5 gm of Ferric Sulfate $\text{Fe}_2(\text{SO}_4)_3$. The resulting mixture was

subjected to ultra-sonication in a bath for a duration of 10 min to enhance component dispersion. The user's text is empty. The sample was stored in a dark environment overnight while being agitated on the shaker. The resultant solution was subjected to centrifugation at a speed of 8000 revolutions per minute for a duration of 10 minutes. Subsequently, it was rinsed twice with deionized distilled water to eliminate any remaining extracellular cells. The powder was then placed in a dark vial for later use after being dried in an oven at a temperature of forty degrees Celsius for an entire night.

Application of parameters (treatments) in the studied area

An area of grand land containing halfa herbs was taken with an area of 3.375 m², which was then split into 6 equal squares depending on the number of treatments used in the studied experiment. It was a square with 75 cm sides and left the space among these squares 10 cm.

The first parameter: Iron oxide nanoparticles (Fe_2O_3 NPs) this parameter has four levels, including

a- Without iron oxide nanoparticles Fe_2O_3 NPs.

b- With iron oxide nanoparticles (Fe_2O_3 NPs) Concentrations 0.5, 1, 1.5 g/ml.

Secondary parameter: The rate of herbicide spraying this parameter has four levels, including

a- Without herbicide.

b- With herbicide in the recommended dose of company.

Through the combinations between the levels of the above parameters, we have six treatments were shown in Table 1.

- Without herbicide - without nanoparticles (negative control).
- Herbicide without nanoparticles (positive control).
- Nanoparticles 1 g/ml - without herbicide.

- Herbicide with nanoparticles 0.5 g/ml.
- Herbicide with nanoparticles 1 g/ml
- Herbicide with nanoparticles 1.5 g/ml.

Table 1. Treatments by adding glyphosate pesticide and iron nanoparticles by spraying

No.	Treatment	Amount of spray	Symbol code
1	Without (Herbicide + Fe ₂ O ₃ NPs)	-	Control negative
2	Herbicide	17 ml	Control positive
3	Fe ₂ O ₃ NPs con. (1) with DDW	10ml+1g	Three
4	Herbicide with Fe ₂ O ₃ NPs con. (0.5)	17ml+0.9g	Four
5	Herbicide with Fe ₂ O ₃ NPs con. (1)	17ml+1.7g	Five
6	Herbicide with Fe ₂ O ₃ NPs con. (1.5)	17ml+2.6g	Six

*The amount recommended by Syngenta, the manufacturer of glyphosate pesticide, % concentration.

The experiment was conducted according to (R.C.B.D.) methodology, with three replications, and applied the specified parameters. A total of 18 experimental units (Table 2).

In this experiment, the following characteristics were studied

- Visual estimation: Phenotypic description of the plant
- Number of leaves on each plant: The number of all leaves on each plant and for all growing plants in each of the treatments was calculated, divided by their number, and recorded.

➤ Number of plants after treatment: The measurements of the number of all growing plants.

➤ Percentage of control %: The control percentage was determined by calculating the ratio of plants growing in each plant. The experimental treatments are determined for each treatment based on the following equation by Ciba-Giegy (1975):

Percentage of control (%) = $\frac{\text{Number of plants in control} - \text{Number of plants in treatment}}{\text{Number of plants in control}} * 100$.

Table 2. Common and trade name, percentage of active ingredient, and usage rate of glyphosate pesticide

Spraying rate	Percentage of active ingredient	Common name	Trade Name
800ml / 30ml/ 1000m ³	36%	Glyphosate	Touchdown S4®

Ultra sonication bath, involves using ultrasonic waves to mix herbicides with nanoparticles. This method can enhance the dispersion of nanoparticles in the herbicide solution. Prepare a mixture of herbicide and nanoparticles in a solvent. Use an ultrasonic bath to apply ultrasonic waves to the mixture for a specified duration 30 min (Yaaqoob, 2022). The ultrasonic energy helps to break up

agglomerates and disperse the nanoparticles evenly.

Results and Discussion

This study collected 42 samples from eight different agriculture regions of Baghdad city to isolate and identify of a group bacteria spread in soil samples. In these samples, a total of 62

bacterial isolates were isolated, different genus belongs to *Enterobacteria*, *Serratia*, *Aeromonas*, *Pantoea*, *Pseudomonas*, *Kocuria*, *Staphylococcus*, *Leclercia*, *Leuconostoc*, *Streptococcus*, *Citrobacter*, *Escherichia*, and *Bacillus*.

Identification of the isolates

The cultural properties of the isolated colonies were determined using the Vitek-2 compact system and biochemical identification.

Biochemical identification

In the present study, a total of 62 isolates were identified from a sample pool consisting of 42 samples. The identification of all the isolates was conducted by the implementation of several biochemical assays, as outlined in Table 3 and Figure 1.

Table 3. Biochemical test results of isolated bacteria

Bacteria	No. of Isolates	percentage% of isolates	Catalase test	Oxidase test	Gram staining	Growth on	
						Mannitol	MacConky
<i>Bacillus spp.</i>	4	6.45	+	+	+	+	N
<i>Staphylococcus hominis</i>	2	3.22	+	-	+	+	N
<i>Streptococcus thortensis</i>	2	3.22	-	-	+	+	N
<i>Leuconostoc pseudomesenteroides</i>	2	3.22	-	-	+	+	N
<i>Kocuria kristinae</i>	2	3.22	+	+	+	+	N
<i>Staphylococcus lentus</i>	2	3.22	+	+	+	+	N
<i>Pseudomonas oryzihabitans</i>	3	4.83	+	-	-	N	+
<i>Enterobacter cloacae complex</i>	10	16.12	+	-	-	N	+
<i>Serratia fonticola</i>	6	9.67	+	-	-	N	+
<i>Aeromonas hydrophila</i>	4	6.45	+	+	-	N	+
<i>Leclercia adecarboxylate</i>	1	1.61	+	-	-	N	+
<i>Pantoea spp.</i>	8	12.9	+	-	-	N	+
<i>Pseudomonas stutzeri</i>	3	4.83	+	+	-	N	+
<i>Pseudomonas mendocina</i>	6	9.67	+	+	-	N	+
<i>Citrobacter freundii</i>	1	1.61	+	-	-	N	+
<i>Escherichia coli</i>	2	3.22	+	-	-	N	+
<i>Pseudomonas aeruginosa</i>	4	6.45	+	+	-	N	+
Total	62						

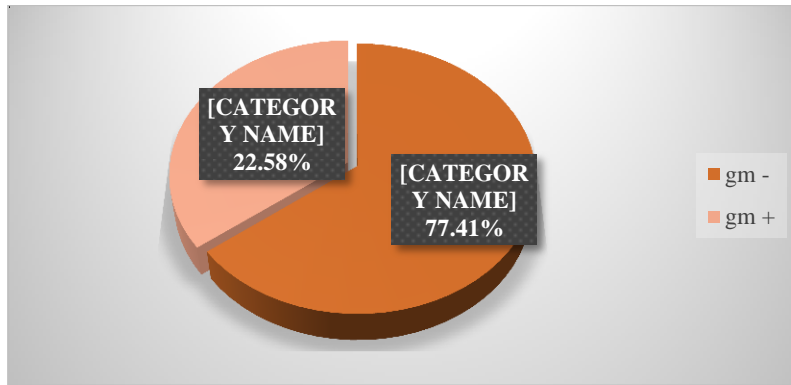


Figure 1. Percentage of Gram-negative and Gram-positive bacteria isolated

Vitek-2 compact system

Based on their physical and biochemical characteristics, and by Vitek-2 compact System these total samples of 62 isolates were

confirmed. The percentage of bacterial isolates from the samples collected from soil after calculating isolate is shown according to Figure 2.

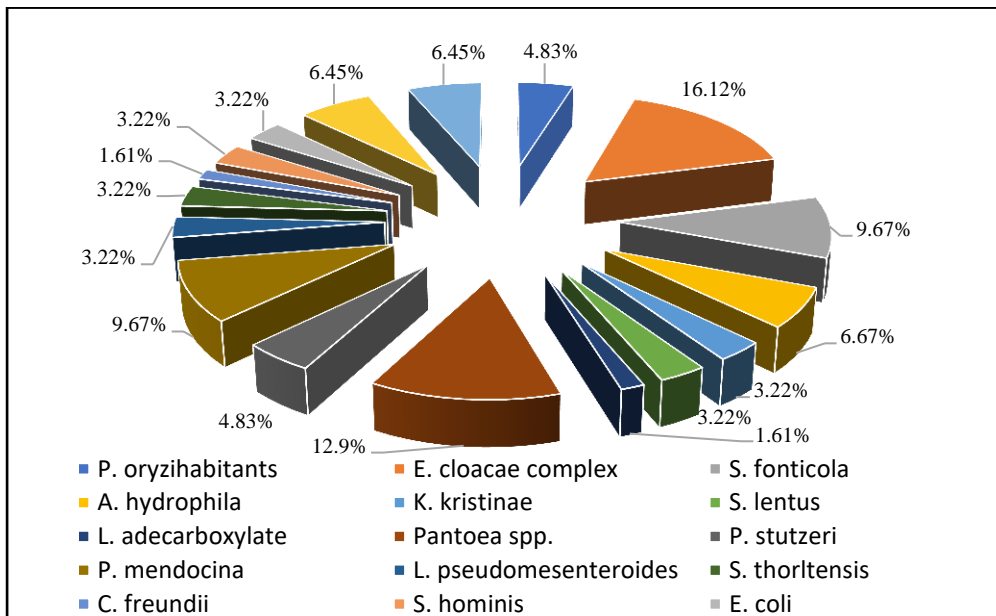


Figure 2. Percentage distribution of isolated bacteria in Baghdad soil

Characterization of extracellular extract of *Serratia fonticola*:

Ultraviolet-visible light (UV-VIS) Scanning Spectrum of extracellular extract of *Serratia fonticola*

Explain the absorption spectra of the used extracellular extract component. The absorption peak was a wavelength of 280 nm. As shown in Figure 3.

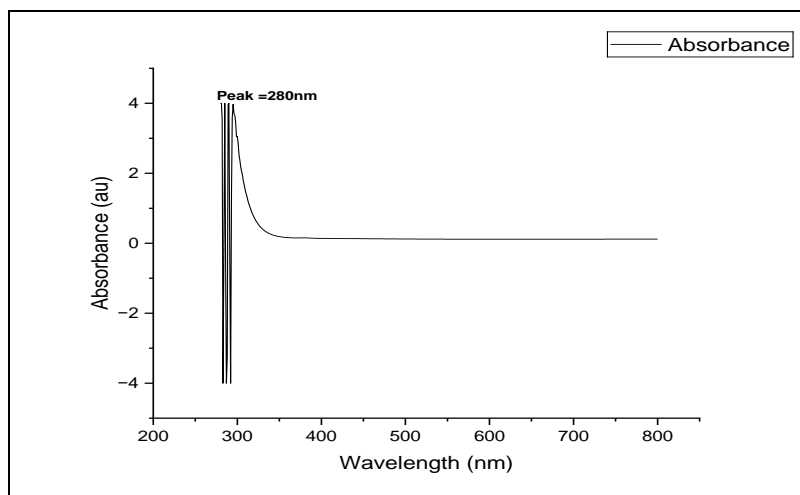


Figure 3. UV-VIS images of extracellular extract of *S. fonticola*

Fourier transform - infrared (FTIR) of extracellular extract of *Serratia fonticola*

The spectrum IR spectrum of the extracellular extract of *Serratia fonticola* lyophilized sample analyzes the spectrum of the six major peaks at 3415.70-3371.34, 3001.03-2935.46, 1573.81, 1415.65, 1124.42-1012.56, and 649.97-430.10 cm^{-1} . A method for determining the bond vibration frequencies of a molecule is FTIR spectrographic analysis. In the extracellular extract of *Serratia fonticola*, the

region between 3415.70 to 3371.34 cm^{-1} has stretching mode of O-H Alcohol, absorption of N-H stretching of Amine salt group is (3001.03-2935.46 cm^{-1}), N-H bonds of amine group is (1573.81 cm^{-1}), O-H bond of Alcohol group is 1415.65, the region between (1124.42-1012.56 cm^{-1}) has group of C-N stretch Amine, and the absorption from 649.97 to 430.10 is due to the represent of the Fe-O band and Fe-O- Fe skeletal frequency, as shown in Table 4.

Table 4. Fourier transform infrared (FT-IR) spectroscopy measurement of extracellular extract of *Serratia fonticola*

	Frequency of Absorption (cm^{-1})	Bonds	Compound class of Functional Groups
Extracellular Extract	3415.70-3371.34	O-H stretch	alcohol
	3001.03-2935.46	N-H stretch	Amine salt
	1573.81	N-H bond	amine
	1415.65	O-H bond	alcohol
	1124.42-1012.56	C-N stretch	amine
	649.97-430.10	Metal oxygen	Metal oxygen

Characterization of Iron oxide Nanoparticles (Fe_2O_3 NPs) from extracellular extract of *Serratia fonticola*

Atomic Force Microscopy (AFM) of Fe_2O_3 NPs:

Atomic force microscopy analysis of Fe_2O_3 NPs surfaces revealed that both 2D and 3D shapes may form. Fe_2O_3 NPs AFM pictures demonstrate the spherical shape of the biosynthesized Fe_2O_3 NPs. The average diameter determined by AFM was 29.21 nm (Table 5).

Table 5. The mean diameter of Fe₂O₃ nanoparticles that were produced from *S. fonticola*

Avg. Diameter:29.21 nm	<=10% Diameter:16.00 nm
<=50% Diameter:26.00 nm	<=90% Diameter:44.00 nm

The examination of ultraviolet-visible light (UV-VIS) spectra the presence of Fe₂O₃ inside the extracellular

The optical properties of the nanoparticles were analyzed with a UV-Visible spectrophotometer. The graphical

representation illustrates the absorbance of the sample within the nanometer range at standard room temperature that to detect the maximum absorption. Absorbance is measured at a wavelength of 359 nm. As shown in Figure 4.

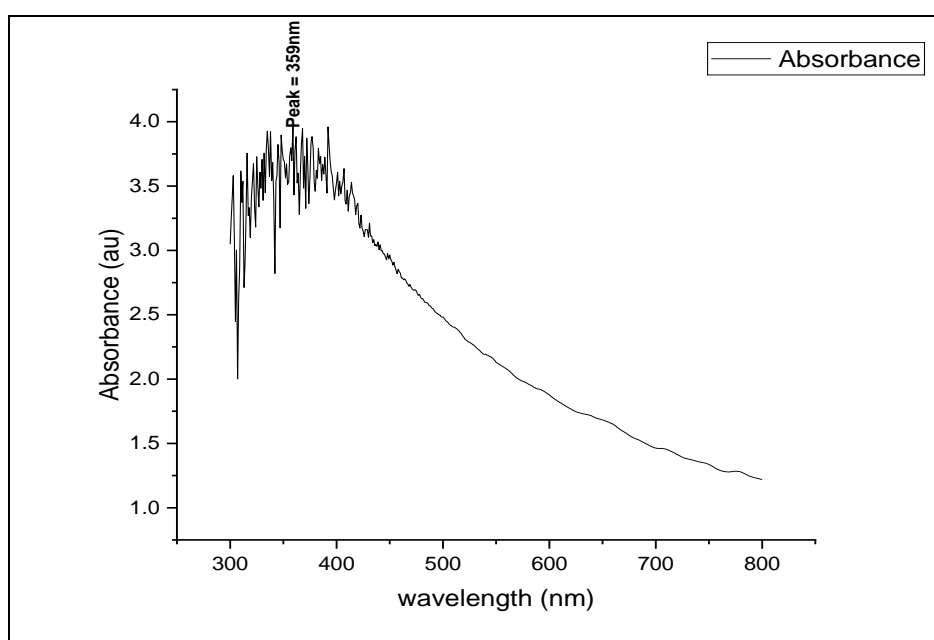


Figure 4. UV-VIS images of Fe₂O₃ NPs synthesized using extracellular extract of *S. fonticola*

Fourier transform infrared (FTIR) analysis of Fe₂O₃ from extracellular

The FTIR spectrum has identified the specific functional groups present in the nanoparticles. Illustrates the absorption spectra of nanoparticles that are produced through biological means using Fourier Transform Infrared Spectroscopy (FTIR). An intense peak at 3434.98cm⁻¹ was visible due to OH stretching mode. The occurrence of the peak properties at

1639.38cm⁻¹ suggested the presence of conjugated acid C=O stretch. The peak at 1552.59cm⁻¹ is due to nitro compound N=O stretch. In the Fe₂O₃ NPs absorption S=O stretch of sulfate compound is (1413.72 cm⁻¹). Present of aliphatic ether compound C-O stretch in 1126.35 cm⁻¹. In 802.33 cm⁻¹ has a C-H bond of 1, 4 disubstituted or 1,2,3,4 tetra substituted. The wide peak at 640.32 to 518.82cm⁻¹ depicted the Fe-O bond and the frequency of the Fe-O-Fe skeletal vibration (Table 6).

Table 6. Fourier transform infrared (FT-IR) spectroscopy measurement of extracellular extract of *S. fonticola*, Fe₂(SO₄)₃, extracellular+ Fe₂(SO₄)₃, and Fe₂O₃ NPs

Extracellular Extract	Frequency of Absorption (cm ⁻¹)	Bonds	Compound class of Functional Groups
Ferric Sulfate	3425.34-3257.55	O-H stretch	Alcohol
	1573.81	N-H bond	amine
	1423.37	O-H bond	Carboxylic acid
	1114.78-1012.56	C-N stretch	amine
	649.97-621.04	Metal Oxygen	Fe ₂ O ₃
Extracellular+Ferric Sulfate	3477.42-3259.47	O-H stretch	Alcohol
	1575.73	N-H bond	amine
	1423.37	O-H bond	Carboxylic acid
	1105.14	C-O stretch	Secondary alcohol
	649.97-621.04	Metal Oxygen	Fe ₂ O ₃
Fe₂O₃ NPs	3434.98	O-H stretch	Alcohol
	1699.17	C=O stretch	Conjugated acid
	1552.59	C=C stretching	Cyclic alkene
	1413.72	S=O stretch	sulfate
	1126.35	C-O stretch	Aliphatic ether
	802.33	C-H bond	1,4 disubstituted or 1,2,3,4 tetra substituted
	640.32-518.82	Metal Oxygen	Fe ₂ O ₃

Field emission scanning electron microscopy analysis (FE-SEM) of Fe₂O₃ from extracellular

The surface morphology of the prepared Fe₂O₃ nanoparticles was revealed through the SEM image. The sample was imaged at a specific magnification 100kx. Focused on (Figure 3). The entire sample exhibits smooth surfaces and a consistent spherical shape

composed of Fe₂O₃ Nano cluster centers. It is worth specifying that the normal nanoparticle size was found to be around (45-55) nm by simple counting and calculations of a number of particles and their sizes, this will confirm the nature of the nanostructure of iron oxides. Where this image appears the delivery of iron oxide NPs with groups. In addition, the FESEM image has approved the formation of iron oxide NPs by *S. fonticola*.

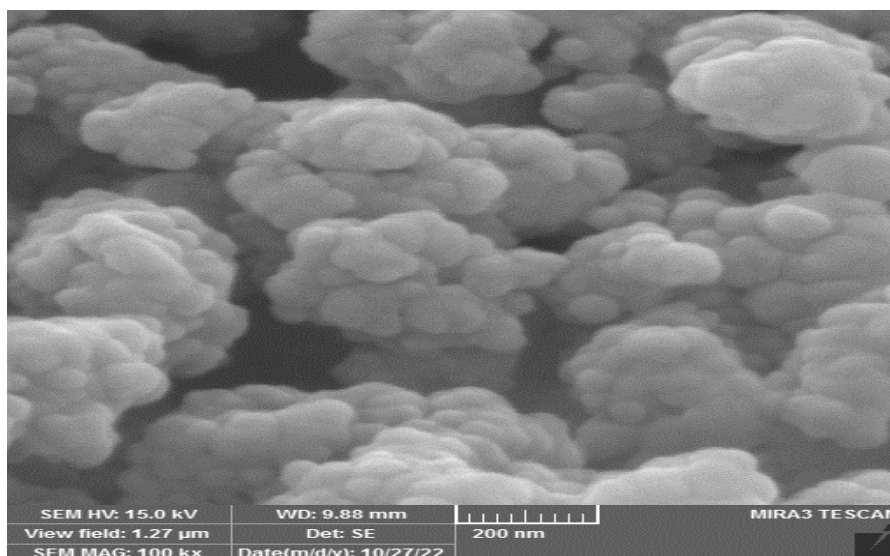


Figure 3. FE-SEM Image of Fe₂O₃ NPs synthesized using extracellular

Application of studied parameters (treatments)

Treatment 1 without Glyphosate herbicide and Fe₂O₃ NPs: this treatment involved no application of herbicide or nanoparticles, this serves as the negative control. The observations noted during the experiment:

- The growth and vertical rise of grass continued with good health
- In the negative control group, the height of halfa grass ranged between 30-40cm. Number of pants and leaves and percentage of control are shown in Table 7.

Treatment 2 with Glyphosate herbicide: this treatment involved the application of only Glyphosate at the recommended dosage, specifying it as the positive control. The observations noted during the experiment:

- After 30 days of treatment, there was a change in the color or health of almost of the grass.
- The color changes and fading were minimal and this is due to resistance to the halfa to Glyphosate. Number of pants and leaves and percentage of control are shown in Table 7.

Treatment 3 Fe₂O₃ NPs con. One g/ml without Glyphosate herbicide: this treatment involved the application of iron oxide nanoparticles (Fe₂O₃) in 1 g/ml. There has been

a slight change in color or health that some of the grass is turning yellow.

Treatment 4 Glyphosate herbicide with Fe₂O₃ NPs con. 0.5 g/ml: this treatment involved the application of Glyphosate at the recommended dosage with Fe₂O₃ NPs con. (0.5) g/ml after 30 days of monitoring.

- That showed a synergistic effect between Glyphosate and 0.5 g/ml of nanoparticles, as it was observed that the color of the grass changed from green to yellow
- As well as wilting and weakness, which indicates that nutrients do not reach them. Number of pants and leaves and percentage of control are shown in Table 7.

Treatment 5 Glyphosate herbicide with Fe₂O₃ NPs con. 1 g/ml: this treatment involved the application of Glyphosate at the recommended dosage with 1 g/ml of Fe₂O₃ NPs. monitoring that showed:

- a synergistic effect between the herbicide and 1 g/ml of prepared nanoparticles, as it was observed that the color of the grass changed from green to yellow.
- As well as wilting and weakness, this indicates that nutrients do not reach them. Number of pants and leaves and percentage of control shown in Table 7.

Treatment 6 Glyphosate herbicide with Fe₂O₃ NPs con. 1.5 g/ml: this treatment involved the application of Glyphosate at the

recommended dosage with 1.5 g/ml of Fe₂O₃ NPs. that showed:

- a synergistic effect that observed the color of the grass changed from green to yellow also
- wilting and weakness
- lack of growth that shows the grass stops growing while the plants around them continue to grow

- Eventually, the grass may show signs of rotting or decay, with the lowest number of plants and leaves and the highest percentage of control and shown in Table 7.

The results of the application of Glyphosate with Fe₂O₃ NPs have been shown in Table 7.

Table 7. The effect of treatments on some vegetative traits of halfa plant

Treatment	Number of leaves/plant	Number of plants/cm ²	Control percentage%
1	72.60 ±3.18 a	12.60 ±0.67 a	0.00 ±0.00 e
2	44.00 ±2.87 c	7.60 ±0.42 b	39.68 ±2.06 c
3	55.66 ±2.91 b	9.30 ±0.61 b	29.38 ±2.17 d
4	38.66 ±1.76 c	6.66 ±0.37 b	47.14 ±3.02 c
5	14.00 ±0.63 d	2.44 ±0.17 c	80.63 ±4.19 b
6	5.33 ±0.29 e	1.26 ±0.08 c	90.00 ±4.52 a
LSD value	8.0278 **	3.164 **	8.157 **

Means having the different letters in the same column differed significantly, ** (P≤0.01).

The number of leaves and control percentage showed a significant difference (P≤0.01) among all treatments. While in a number of plants, there was no significant difference (P≤0.01) among treatments 2, 3 and 4, in addition to no significant difference (P≤0.01) between treatments 5 and 6, while treatment 1 showed significant differences (P≤0.01) with all the treatments.

The results show that treatments with Fe₂O₃ NPs have significant differences (P≤0.01) that lead to the method of preparation and addition of Fe₂O₃ NPs and compound (herbicide) is the suspension method, and from the results, we note that increasing the concentration of Nano of iron with the glyphosate will increase the effect of killing or controlling the weed.

Nano suspension of glyphosate cause a synergy to occur between the active ingredient of the glyphosate that affects a part of the grass and the effect of the nanoparticle, leading to an increase in the

targeted part of the grass, increased solubility of water-insoluble Nano suspension of glyphosate, eliminate the toxic organic solvents, and would gradually substitute the conventionally EC products (Liu *et al.*, 2011; Zhang *et al.*, 2008; Rabinow, 2004). In addition, the most of crop leaf surfaces are highly hydrophobic which inhibits liquid deposition and reduces the spray drift and run-off loss on the hydrophobic foliage (Whitehouse and Rannard, 2010). Pesticide Nano formulations increase the adhesion and deposition of droplets on the leaves through leaf-affinity modification.

Conclusions

The study successfully bio-synthesized iron oxide nanoparticles using an extracellular extract from *Serratia fonticola*. This green synthesis approach is a significant contribution to the field of nanotechnology, particularly for agricultural applications. This study demonstrated the effectiveness of these

nanoparticles in conjunction with Glyphosate herbicide on the management of Cogon grass. This could potentially offer a new method for controlling this invasive species. The research findings indicate a synergistic effect between the iron oxide nanoparticles and Glyphosate herbicide. This synergy led to enhanced herbicidal activity, as evidenced by changes in grass color and reduced growth. The best concentration of Fe₂O₃ was 1.5 g MI⁻¹ with Glyphosate, and the wilting result appeared after treatments. With 90% of percentage control. For environmental implications: The use of biologically synthesized nanoparticles for herbicidal purposes could be a more environmentally friendly alternative to traditional chemical methods. This could have significant implications for sustainable agricultural practices.

Conflict of Interest

The authors declare no conflict of interest.

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