Original Article

Modeling phenol biodegradation with *Pantoea agglomerans* as plant-growth-promoting bacteria

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ABSTRACT

In the current investigation, the potential of using the bacterial species Pantoea agglomerans for phenol biodegradation was evaluated. The inhibitory effects of various doses of phenol (ranging from 200 to 1200 ppm) on the growth of P. agglomerans and the biodegradation rate were evaluated. The effects of temperature and pH were also investigated in order to determine the ideal parameters for maximum phenol biodegradation. Additionally, the ideal parameters for growth rate and beginning phenol content were determined using mathematical modeling employing modified Gompertz and Haldane models. The half-saturation coefficient (Ks) for Haldane and the maximum specific growth rate (max) for phenol-dependent growth kinetics (Ki) has been calculated to be 480.23 mg/L, 188.17 mg/L, and 0.887 mg/L, respectively. The Haldane equation can be applied to empirical data because of its tiny sum of squared error (SSR), which is 1.39 x 10-3. Additionally, the reformulated Gombertz model effectively predicts trends in the degradation of phenol. The starting amount of phenol impacted both the degradation rate and the period of lag time, and both of them raised further as the phenol level got higher. A 28°C incubation temperature and a pH of 7.0 were determined to be the ideal conditions for Pantoea agglomerans to grow and degrade phenol. The GC-MS data showed that many phenol derivatives were produced due to phenol degradation, such as catechol, 2-hydroxymucconic semialdehyde, and pyruvate. This may provide strong evidence that the degradation of phenol by P. agglomerans occurred via a meta-pathway.

Keywords: Phenol, Biodegradation, Pantoea agglomerans, Meta-pathway, Catechol, Mathematical modeling

Introduction

All forms of life are dependent, in a fundamental way, on the utilization of chemicals by living cells as sources of carbon or energy. The large variety of organic species that humans have produced has resulted in environmental problems because they are resistant to mineralization by any living organism, even though it is now widely acknowledged that living cells have evolved to be able to assimilate the natural chemical compounds found on Earth [1-4]. This has led to an increase in the amount of pollution in the environment. Starting ingredients typically consist of phenol and various substituted phenols. These materials are considered to be waste by-products that result from the production of agricultural and industrial products [5-8]. Phenol can be harmful to certain aquatic organisms at doses in the low mg/l range, and it can cause abnormalities in the taste and odor of drinking water at concentrations that are much lower than that [9-11]. Because of the widespread application of phenols in the United States as well as the potential toxicity of these chemicals, the Environmental Protection Agency of the

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United States has placed them on the data of highest-priority pollutants [12-14].

There are serious drawbacks when removing phenol from the environment using techniques such as abiotic treatment, solvent extraction, chemical oxidation, burning, and adsorption [15-17]. These drawbacks include economic concerns as well as the development of harmful byproducts. In most cases, biodegradation is chosen since it results in lower costs and full mineralization of the waste. When it comes to biodegradation, there is a great deal of debate on whether natural or genetically engineered microorganisms (GEM) should be used. Because of the possibility of unanticipated effects on the surrounding ecosystem, most government agencies are reticent to let GEMs be released into the environment [3, 12, 18]. There is a significant amount of interest in the process of isolating microorganisms that can flourish in environments with high concentrations of aromatic chemicals [19-22], such as the phenol molecule that was investigated in this research. In contrast, there is no evidence that Pantoea agglomerans uses phenol or any other aromatic chemicals as its sole source of carbon and energy. The way that catechol is broken down, the availability of nutrients (sources of carbon and nitrogen), the presence or absence of toxins, and environmental variables (like temperature) have all been shown to have an impact on the growth of bacteria on phenol [20, 23-25]. A facultatively anaerobic strain called Pantoea agglomerans has been connected to the biodegradation of vaseline, toluene, and kerosene [26-28]. The development and dispersal of Pantoea agglomerans are affected by environmental factors such as the severity of winter chilling, the amount of exposure to sunlight, and the quality of airflow. Pantoea agglomerans are able to produce antibiotic compounds, and these substances are toxic to the bacterium that causes fire blight. According to Rezzonico *et al*. [29], Pantoea agglomerans may be effective for biologically reducing fire blight through strategies such as changing the environment or excluding other organisms from the area (competition). It's commonly accepted that bacteria have evolved to eat earth's inherent organic components. The vast array of organic forms that humans have generated has created environmental challenges because they are completely resistant to being mineralized by any type of bacteria. Human actions are the root causes of these problems. Environmentally hazardous issues have consequently surfaced. In the present study, the biodegradability of phenol was investigated using the isolated growth-promoting bacteria Pantoea agglomerans. One of the other objectives of the study was to determine the optimal pH and incubation temperature for P. agglomerans to biodegrade phenol.

Materials and Methods

Bacterial strain and culture condition

Pantoea agglomerans were used in this study. This strain was isolated from Al-Ghweiler Agricultural Station, Karak, Jordan. The identification was performed using 16S rRNA, and the nucleotide accession number (JOR33 MN083295) was given for this species by the Genebank. The strain is housed in the microbial collection at MIRRI-It ENEA in Italy.

Wolfe's mineral solution and phenol-free mineral medium were prepared according to the standard instructions. The concentrations, volumes, and culture conditions in these media were prepared based on our previous investigations [30]. The tryptophan-containing medium was prepared as follows: L-tryptophan (0.5 g yeast, 0.5 g peptone, 0.5 g casein hydrolysate, 0.5 g glucose, 0.5 g soluble starch, 0.3 g $K_2HPO_4 \cdot 3H_2O$, 0.024 g $MgSO_4$, 0.3 g sodium pyruvate, and 1 L water, pH 7.0) [31]. Other media used in this study were prepared according to the manufacturer's instructions.

Growth-Enhancing parameters

Indole-3-acetic acid (IAA)

The production of indole-3-acetic acid (IAA) by P. agglomerans was assessed using a tryptophan-containing mixture and Salkowski's reagent. After three days of incubation at 30 degrees Celsius, the development of a pink or red tint indicated the formation of IAA [31].

Phosphate solubilization

PKO medium was used to determine the ability of *P. agglomerans* to solubilize phosphate. The culture was incubated at 30C for 7 days and monitored daily for the formation of a transparent zone around the colonies [32].

Siderophore production

Chrome azurol S (CAS) agar was used to qualitatively assess the siderophore synthesis method. The production of siderophore by *P. agglomerans* was indicated as a change in the medium color from blue to green [32].

Nitrogen fixation

A semi-solid New Fabian medium was used to assess P. agglomerans' capacity to fix nitrogen. In this test, a portion of *P. agglomerans* (absorbance 0.5 at 600nm) suspended in phosphate buffer saline was added to 4 mL New Fabian medium. The prepared culture was incubated for 72h at 28°C [33]. The studied bacteria was considered an N2 fixative as it could grow as a sub-surface pellicle on an NFb medium.

Phenol as a carbon source and phenol degradation

This test was performed using mineral media. In brief, a portion of *P. agglomerans* culture reaching the log phase was centrifuged and resuspended twice in mineral media to get OD600 nm of 0.2. Then, the prepared bacterial suspension was cultured in a mineral medium containing phenol (700 ppm). The culture was incubated for 96h. During the incubation period, a portion of the culture was removed and measured at 12h intervals using a

spectrophotometer at 600nm. Mineral media of 700 ppm phenol without bacteria was used as a positive control.

4-aminoantipyrine colorimetric method to determine phenol concentration

This test was performed according to [34]. The phenol concentrations in the cultured media prepared as shown in the previous section were measured. At room temperature, a solution containing 0.5 N ammonium hydroxide, 2% w/v 4-aminoantipyrine, and 8% w/v potassium ferricyanide was incubated. The OD510 nm was taken after 15 minutes and utilized to create the phenol standard curve and linear equation. To prevent mistakes arising from widely varied lag period lengths, the concentration of residual phenol converted during the first 24 hours was determined. Additionally, an attempt was made to estimate the time required to complete biodegradation or the point at which biodegradation stops to take place [35].

Optimization of incubation conditions (pH and temperature) for optimum phenol biodegradation

Mineral media containing phenol of 700 ppm was prepared in 4 sets. Then, the pH of each media was adjusted to produce media with pH of 5.5, 7.0, 8.0, and 9.0. Likewise, the previously used testing culture was incubated at various incubation temperatures, which include 25, 28, 33, and 37°C, with the objective to study the impact of the temperature on phenol degradation.

Mechanism of biodegradation

Gas chromatography-mass spectrometry analysis

GCMS analysis was performed to determine phenol and phenol derivatives in inoculated mineral media containing 700 ppm at the end of the incubation period (96h). in brief, the samples were prepared by adding 10% CuSO4 to the cultured media. Then, the contents of the samples were extracted using DCM (dichloromethane). After removing the solvent, the samples were solubilized in methanol and subjected to Varian Chrompack CP-3800 GC-MS-200 (Saturn) equipped with a DP-5 column (30 m 0.25 mm i.d., 0.25 m film thicknesses). The samples were analyzed using a mobile phase of Helium gas at a flow rate of 1 mL per minute. The ionization voltage and the MS temperature were set to 70 eV and 180 °C, respectively. The temperature of the column was programmed to reach 270 °C at a rate of 3 °C/min and an isothermal program of 60 °C for 1 minute was used. The contents identification was performed based on a comparison with standard compounds (C8-C20), retention time, and mass spectra of the published data of the NIST database.

High-performance liquid chromatography (HPLC) analysis

To measure the amount of phenol and phenol derivatives in inoculated mineral media at 700 ppm and at various time intervals including 0, 12, 24, 48, 72, and 96 hr, HPLC analysis was conducted. In brief, samples were analyzed using an HPLC/UV-Vis detector (Shimadzu, LC-10A, Tokyo, Japan) and a Luna C18 column (4.6 250 mm, 5 m, 100Ao). A mobile phase of acetic acid (A) and acetonitrile: methanol (1:1) (B) as follows: 0.1-1 min, 95 percent A; 1-6 min, 50 percent A; 6-10 min, 5 percent A. the eluted compounds was determined using a UV detector at 280 nm. Also, phenol and catechol were used in this test as standard compounds. Their retention times were determined and compared with the corresponding compounds in the tested samples.

Mathematical modeling

The growth pattern in a batch culture, which includes exponential phase and stationary phases, is typically described by the logistic equation. In differential form, the logistic equation is stated as follows:

$$\frac{dX}{dt} = \mu_m X\left(1 - \frac{X}{X_m}\right) \quad (1)$$

Where μ_m is the highest growth rate (hr^{-1}) viable in a certain environment and X_m is the number of bacteria that can be present in that environment at their highest concentration.
By integrating the logistic equation, the following cell concentration formula is created:

$$X = \frac{X_0 e^{\mu_m t}}{1 - \left(\frac{X}{X_0}\right)(1 - e^{\mu_m t})} \quad (2)$$

Where X_0 is the number of the bacterial cell at time 0 min.

Haldane equation has been applied here to find the correlation between the substrate concentration (phenol) S and the rate of bacterial growth μ. Hence, the inhibitory effect of using phenol as a substrate at high concentrations can be indicated:

$$\mu = \frac{\mu_{\max} s}{K_s + S + \frac{S^2}{K_l}} \quad (3)$$

where is the half-saturation coefficient (ppm), the inhibition coefficient of growth kinetics (ppm), and the μ_{maxs} (specific growth rate (hr^{-1})).

The logistic equation has been fitted to the bacterial growth patterns at different phenol concentrations by the use of nonlinear regression. The ideal model parameters were found by minimizing the sum of squared errors (SSR) with Microsoft Excel 2007's Solver add-in. This process yielded the model fitting

parameters. The profiles of the logistic model and the experimental data at different initial amounts of phenol are shown in **Figure 1**.

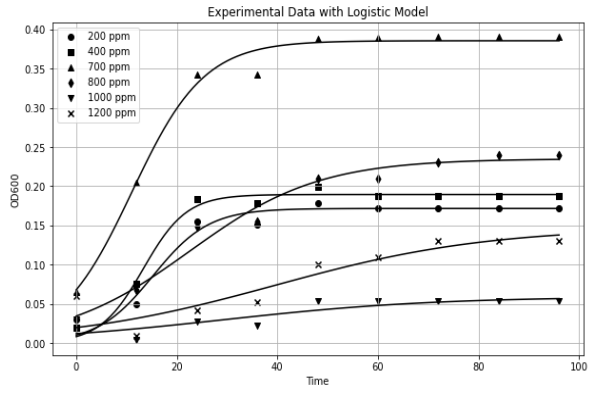

Figure 1. Growth curves of *P. agglomerans*. At different initial concentrations of phenol

Results and Discussion

Growth-enhancing parameters

P. agglomerans' phosphate solubilizing capacity, indole-acetic acid synthesis, nitrogen fixation, and siderophore production were all studied as potential growth promoters. Except for the fact that its indole-acetic-acid production was fluctuating, it exhibited all of the aforementioned characteristics **(Table 1)**.

Table 1. Characteristics of P. agglomerans obtained from the soil of the Al-Ghwer Agriculture Station that promote plant growth

Parameter	Results
indole-acetic acid	+/-
Siderophores	++
Phosphate solubilization	++
Nitrogen fixation	+

Biodegradation of phenol

In this study, *P. agglomerans* was isolated from agricultural soil and cultured on a minimal medium supplemented with phenol as the sole source of carbon and energy to investigate its kinetics. For the first time, *P. agglomerans*, a newly discovered plant growth-promoting bacterium (PGPB), was used to try phenol biodegradation. The rate of phenol biodegradation increased with increasing phenol concentrations. This steady rise in the rate of biodegradation was noted up to a phenol concentration of 700 parts per million. The pace at which phenol was degraded was unaffected by an increase in phenol concentrations at levels higher than 700 ppm. In parallel, negative controls included heat-killed phenol-containing culture broth and uninoculated minimum media broth, both of which contained different concentrations of phenol showing the absence of phenol biodegradation evidence. These results may suggest that the tested strain was the cause of the phenol's biodegradation. *P. agglomerans* could utilize large amounts of phenol as a major source of energy as well as carbon

Phenol biodegradation growth kinetics

The primary substrate, phenol, was used in six distinct amounts **(Figure 1)**. Nonlinear regression was used to fit the experimental bacterial growth patterns at various starting concentrations of the substrate (phenol) to the logistic equation. The model fitting parameters were found by lowering the sum of squared error (SSR) using the Solver add-in for Microsoft Excel 2007. **Figure 1** illustrates the characteristics of logistic regression as well as the experimental outcomes across different phenol concentrations. As can be observed, the biomass growth predicted the maximal stationary population density at a phenol quantity of 700 ppm. The greatest growth rate of the bacteria was achieved at a concentration equal to 400 ppm. At the highest concentrations tested, 1000 and 1200 ppm, the growth was indicated by a poor growth rate and a relatively small, stationary population size **(Table 2)**.

Table 2. Parameters of the growth kinetics of phenol biodegradation by P. agglomerans in a logistic manner

Phenol Concentration (ppm)	$\mu_m\mu_m$ (hr^{-1})	X_mX_m (OD_{600})	SSR
200.0	0.181	0.172	1.0×10^{-2}
400.0	0.232	0.189	4.35×10^{-3}
700.0	0.139	0.386	1.2×10^{-3}
800.0	0.078	0.235	1.5×10^{-3}
1000.0	0.047	0.059	8.48×10^{-4}
1200.0	0.047	0.147	2.69×10^{-5}

The specific growth rate (μ) can be calculated by plotting the bacteria growth rate at the log phase for starting phenol concentrations versus the bacteria growth rate at the different incubation intervals. The specific growth rate was calculated using:

$$\left(\frac{lnX}{X_0}\right)^2 \qquad (4)$$

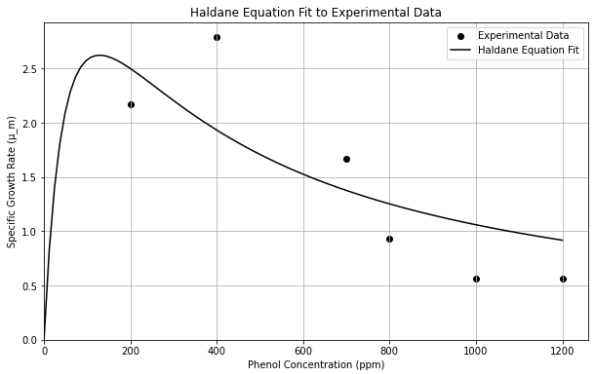

Figure 2. *P. agglomerans* strain experimental batch growth data were fitted using a Haldane growth kinetic model.

A growth kinetics pattern for the inhibitory effect of the substrate using the Haldane equation is depicted in **Figure 2**. To

determine the Haldane parameters, non-linear regression based on SSR reduction was used. These parameters include the coefficient of growth inhibitory, the coefficient of half-saturation, and the highest rate of growth were found to be 480.23 ppm, 188.17 ppm, and 0.887 ppm respectively. The data was found to fit the Haldane equation at SSR of 1.39×10^{-3}.

The specific growth rate increases with increasing phenol concentration up to around 200 ppm, at which point it begins to decrease as starting phenol quantity increases, according to the results. The greatest specific growth rate determined by the Haldane equation in this investigation is in close proximity to the values discovered in other investigations concerning the breakdown of phenol by bacteria [36]. The half-saturation coefficient from the Haldane model, which was calculated as 188 ppm, shows that the concentration of phenol must remain minimal in order to achieve a specific growth rate equivalent to half the highest specific growth rate. In Haldane's growth kinetics, the reaction of biomass to agents that slow growth is measured by the inhibition coefficient. The fact that the inhibitory constant is 480 ppm shows how well phenol stops biomass from growing. To mimic how phenol breaks down, the revised Gombertz model was used:

$$S = S_0\left\{1 - \exp\left\{-exp\left[\frac{R_m}{S_0}e(\lambda - t) + 1\right]\right\}\right\} \quad (5)$$

The biodegradation patterns of phenol match the modified Gombertz model **(Table 3)**. When there is less phenol to start with, the phenol biodegradation rate continues as the amount of phenol increases. Most phenol was degraded at a rate of 11.91 ppm/h when the concentration of the phenol was 700 ppm **(Figure 3)**. So, 700 ppm was selected for more testing based on the growth pattern and the amount of phenol that was removed. Accordingly, the effect of phenol concentrations, the temperature of the incubation, and the pH of the growth medium on the biodegradation of phenol was examined.

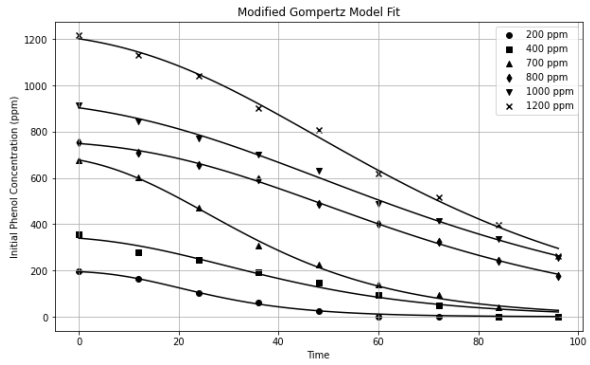

Figure 3. Growth pattern of *P. agglomerans using modifi*ed Gompertz model at different concentrations of phenol.

Table 3. Gompertz model Parameters were used on the six initial concentrations of phenol.

Phenol Concentration (ppm)	R_mR_m (mg/L.hr)	$\lambda\,\lambda$ (hr)
200	5.15	5.88
400	5.44	5.51
700	11.91	4.04
800	7.79	13.21
1000	8.21	6.41
1200	12.20	10.29

Optimization conditions

Incubation temperature

In order to find the optimum temperature, the effect of four different incubation temperatures on the rate of phenol biodegradation was examined The results showed that the optimum temperature at which the maximum rate of biodegradation takes place is 28 °C. An increase or decrease in temperature far from the optimum leads to a decrease in the phenol biodegradation rate data not shown.

PH

The effect of culture pH on the rate of phenol biodegradation was evaluated The pH of 7.0 was found to be the optimum pH at which the maximum rate of phenol biodegradation was observed. Above or below this point, a remarkable decrease in the biodegradation rate was observed data not shown.

GC-MS analysis

GCMS was employed in this study to obtain insight into the biological degradation of phenol. Both phenol and catechol were analyzed as a standard. Generally, the GC-MS chromatograms results for the tested sample under investigation, (data not shown) show more than 14 peaks with various retention times. The catechol and/or phenol metabolism and condensed products are revealed by the GC-MS analysis of those significant peaks and were recognized using the NIST mass spectral database. Regardless of the culture media compositions indicated by the GCMS, phenol derivatives compositions such as catechol, acetaldehyde, and propanal **(Table 4)**. In addition, the results showed the creation of 2-vinylfuran, [1,1'-biphenyl]-2,3-diol, [1,1'-biphenyl]-2,2',3,3'-tetrol, [11,21:24,31-terphenyl]-13,14,34-triol, 4-phenoxyphenol and 3-(1-hydroxyethyl) benzene-1,2-diol.

Table 4. Phenol derivatives composition using GCMS analysis.

	Retention time (min)	Compounds
1.	4.749	2-vinylfuran
2.	10.613	4-(5-hydroxy-4-methylhexan-3-yl)phenol

3.	11.8	Catechol
4.	12.048	3-(4-hydroxyphenyl)-2- oxopropanoic acid
5.	12.243	propanal
6.	15.267	4-phenoxyphenol
7.	15.852	1,1'-[(*1E*)-but-1-ene-1,3-diyl] dibenzene
8.	16.730	3-(3,4-dihydroxyphenyl)-2-hydroxypropanoic acid
9.	17.262	4-(1-hydroxypropyl) phenol
10.	17.636	1,1'-biphenyl]-2,3-diol
11.	18.296	3,3'-(1-hydroxypropane-1,1-diyl)di(benzene-1,2-diol)
12.	18.695	3-(1-hydroxyethyl) benzene-1,2-diol
13.	19.394	$1^{1},2^{1}:2^{4},3^{1}$-terphenyl]-$1^{3},1^{4},3^{4}$-triol
14.	21.765	(*3E*)-4-(3,4-dihydroxyphenyl)-2-oxopent-3-enoic acid (12)

HPLC

HPLC analysis was performed to determine the phenol concentrations and derivatives in mineral media containing 700 ppm phenol at 0, 12, 24, 48, 72, and 96-hour time intervals. A remarkable reduction in phenol concentrations was observed (data not shown). The concentration of phenol decreases as the incubation period increases, and at the end of the incubation period (96 h), a complete biodegradation of phenol was noted. Besides, catechol, as an early derivative formed due to phenol degradation, was significantly increased after 12 h of incubation. However, the concentration of catechol decreased gradually and reached 0 ppm at the end of the incubation period (96 h).

In this study, the bacterium *P. agglomerans* has been isolated from Jordan's Al-Ghweiler Agricultural Station and used to biodegrade phenol. Using 16S rDNA, this bacterium was identified. The growth kinetics and biodegradation of phenol were evaluated using *P. agglomerans*. This research has shown that *P. agglomerans* can create substances that stimulate plant growth. Therefore, these isolates may be used as biofertilizers or bioenhancers for plants [30]. Under iron-limiting conditions, bacteria secrete iron-chelating molecules with a low molecular weight called siderophores. Phosphorus, like many other essential elements, can be found in the soil in an insoluble or fixed form, making it unavailable for plant growth unless its mobility in the soil is increased. Siderophore-producing microorganisms promote plant growth by making iron available to plants and depriving plant pathogens of iron, which stunts pathogen growth. It has been shown that *P. agglomerans* produces indole acetic acid (IAA), the primary auxin that promotes the development of new shoots and roots. The nitrogen-fixation characteristics are also presented in this isolate which is controlled by the nitrogenase enzyme complex, expressed by the nif gene [3].

A minimum medium containing phenol was utilized to confirm that phenol might be the main source of carbon and energy for these bacteria. If biomass is generated and organisms flourish, this is most likely due to the organisms using phenol as a substrate [9, 15]. Phenol-containing minimum media broth (MMB) and heat-killed phenol-containing culture broth were employed as negative controls to measure the degradation of phenol. The findings indicated that there was no sign of phenol biodegradation, indicating that *P. agglomerans* metabolisms were responsible for phenol degradation. Several physiological conditions may impede the bacteria's ability to biodegrade phenol. The toxicity of phenol, the availability of carbon and energy, the environmental conditions (such as temperature and pH), and the abundance of micro- and macro-elements are all important. The incubation temperature and medium pH were evaluated since *P. agglomerans* employs phenol as a carbon and energy source in this study. The results demonstrated that the incubation temperature significantly affected the growth of bacteria and the degree of phenol degradation. At 28 °C, the optimal temperature for *P. agglomerans* cells, it appears that phenol biodegradation takes place naturally at room temperature. It has been demonstrated that temperature has a significant effect on phenol metabolism [36, 37]. Since it has been found that 28°C is the optimal environment for phenol degradation, or probably due to the outcome of temperature-related changes in enzyme activity [38, 39]. According to certain findings, temperature may have an equivalent or higher impact on the breakdown of different organic compounds than nutrient availability [40, 41].

The influence of pH on the concentration of phenol was investigated in an uninoculated culture in order to ascertain if the phenol reduction was caused by a chemical process or something else. It was found that the amount of phenol in the uninoculated culture is significantly influenced by pH. Seven was the ideal pH for biomass growth rates and phenol biodegradation. The medium's pH significantly affects the capacity of microorganisms to degrade by influencing their growth [42]. Certainly, the enzymes play an essential part in this catabolism process, and their maximum activity occurs at a pH of 7.0. As previously mentioned, pH serves a crucial function in the biodegradation of these diverse aromatic compounds. despite this, the optimal pH varies based on the type of microorganisms [42]. For instance, *Arthrobacter*'s optimal pH for the degradation of 4-CBA was 6.8 [43], a value that was comparable to *Klebsiella oxytoca*'s optimal pH for the degradation of phenol [20, 44], whereas *Halomonas campisalis* exhibited significant degradation ability toward phenol and catechol at pH ranges between 8 and 11 [45].

Similar to other examined bacterial strains such as *Pseudomonas sp*. BZD-33 [46], *Pseudomonas aeruginosa* PDM [47], and

Rhodococcus UKMP-5M [48], *P. agglomerans* showed considerable phenol biodegradation at pH 7. Various aromatic compounds have been observed to be degraded by microorganisms possessing oxygenase enzymes. Numerous studies have demonstrated that the proper amount of carbon and nitrogen sources must be supplied in order to maximize the rate of degradation [26, 49]. Since phenol was the only carbon source, the high concentration of phenol-degrading enzymes may have contributed to the quick rate of phenol breakdown by *P. agglomerans* cells. Since microorganisms consume this substrate quickly, phenol toxicity is also reduced [26, 50].

In this investigation, GCMS was utilized to understand the biodegradation of phenol by *P. agglomerans. P. agglomerans* had the ability to convert phenol to catechol, as it was also proved by HPLC, and to convert catechol to 2-hydroxymucconic semialdehyde, suggesting that *P. agglomerans* is employing a meta-pathway (data not shown). The meta route led to the manufacture of pyruvate and acetaldehyde, which eventually broke down into CO_2 and H_2O. Additionally, every precursor molecule is created along the degradation pathway through several oxidation processes. Yet, these molecules were effectively consumed by the bacteria, which allowed us to find them in our analysis. The production of 2-vinylfuran demonstrated that phenol degradation happened via a reverse Diels-Alder reaction. Four compounds were identified when phenol and catechol were combined, namely [1,1'-biphenyl]-2,3-diol, [1,1'-biphenyl]-2,2',3,3'-tetrol, [11,21:24,31-terphenyl]-13,14,34-triol and 4-phenoxyphenol. On the other hand, the elution of 3-(1-hydroxyethyl) benzene-1,2-diol appears to be due to the condensation of phenol and/or catechol with acetaldehyde. Also, the biodegradation of the sample yields propanal. A condensation of propanal occurred with what appears to be phenol and/or catechol to form a 4-(1-hydroxypropyl) phenol and 3,3'-(1-hydroxypropane-1,1-diyl) di (benzene-1,2-diol).

Conclusion

The bacterium *P. agglomerans*, recognized for its ability to stimulate plant growth, was used to study the growth kinetics and biodegradation of phenol. The Haldane equation is highly compatible with actual data. The improved Gombertz model accurately predicts phenol biodegradation trends. The experiment identified the ability of *P. agglomerans* to break down phenol and promote synchronized plant growth. Under conditions of phenol-induced stress, this approach may facilitate the process of rhizoremediation and contribute to the preservation of the yield of crops.

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Ethics statement: None

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