The intensified effect of nitrogen removal properties using *Pseudomonas fulva* K3 and MgBC for the weathered crust rare earth wastewater treatment

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**Abstract:** A new bacteria named *Pseudomonas fulva* K3 (P. fulva) strain was isolated from the surroundings of weathered crust rare earth tailing with efficient NH$_4^+$-N removal ability via heterotrophic nitrification and aerobic denitrification. The nitrogen removal properties could be intensified by the synergistic effect between as-prepared magnesium-modified biochar (MgBC) and P. fulva strain. The results show that P. fulva exhibited a rod-shaped morphology and NH$_4^+$-N can be completely biodegraded under the optimal conditions of pH=7.0~8.0, temperature 30 $^\circ$C and initial NH$_4^+$-N concentration of 100 ~150 mg/L. The NH$_4^+$-N tolerant concentration for P. fulva was determined to be 300 mg/L. The magnesium-modified biochar (MgBC) worked as an adsorbent of NH$_4^+$-N. The kinetics and isotherm model for adsorption could be described by the pseudo-second-order kinetic and Freundlich model, respectively. The XPS results showed that NH$_4^+$-N was mainly adsorbed on the surface by chemical adsorption. Furthermore, the P. fulva could be immobilized on MgBC due to its large surface area, adjusting the concentration of NH$_4^+$-N to a proper range for the growth of P. fulva by adsorption and desorption equilibrium, and leading to the intensified effect on nitrogen removal. The total nitrogen removal efficiency of the eluted weathered crust rare earth tailing reached 84.7 % in MgBC + P. fulva system.

**Keywords:** weathered crust rare earth wastewater, nitrogen removal, pseudomonas fulva K3, magnesium-modified biochar, intensified effect

1. Introduction

Weathered crust rare earth ore is a crucial metal resource in the world, which contains large amounts of middle and heavy rare earth elements (He et al., 2016; Zhang et al., 2016a). More and more attention has been paid to the exploration and environmental protection due to its important role in economics and industries, such as luminescent materials, permanent magnet materials, rechargeable batteries, petrochemicals and metallurgical machinery et al. (Dutta et al., 2016). The rare earth elements are existed as hydroxyl aqueous ions adsorbed on the clay minerals, resulting in the ion-exchange method as its recovery technology in industry (Zhou et al., 2019). And ammonium salts are widely used as leaching agents for the extraction of rare earth elements by in-site leaching process (Feng et al., 2018; Wu et al., 2020; Zhao et al., 2017). Quite amounts of ammonia nitrogen (NH$_4^+$-N) is still remained in the ore body after the extraction process and it can be easily eluted by the rain, flowing into surface water and causing eutrophication (Li et al., 2017). It is of great significance to find an eco-friendly and effective method for the removal of NH$_4^+$-N wastewater eluted from weathered crust rare earth tailing.

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Several methodologies including blow-off, adsorption (Kizito et al., 2015), chemical precipitation (Huang et al., 2010), and biological technique (Ma et al., 2017b) have been mainly developed for the treatment of NH$_4^{+}$-N wastewater. Compared with the physical and chemical removal methods, biological technique has the advantages of low cost, high security, and environmental friendliness (Zhang et al., 2019b). The existing conventional biological processes, such as A/O, A2/O, SBR, and MBR, are established on activated sludge and biofilm (Ahmad et al., 2017; Gong et al., 2018; Luo et al., 2017; Shams et al., 2018). NH$_4^{+}$-N is firstly oxidized to NO$_3^{-}$-N and NO$_3^{-}$-N by autotrophic ammonia oxidizing bacteria (AOB) and nitrite oxidizing bacteria (NOB), respectively (Zhang et al., 2019a). Then the NO$_2^{-}$-N and NO$_3^{-}$-N are reduced to N$_2$ by heterotrophic denitrifying bacteria under anaerobic condition. However, the key factors limiting the industrial application of conventional biological method are the low nitrogen removal speed, tedious procedures and high energy consumption. Given those disadvantages of conventional biologic process, some new techniques are proposed to reduce the route and cost, such as shot cut nitrification-denitrification (Hou et al., 2017), anaerobic ammonium oxidation (Kosugi et al., 2020; Jia et al. 2017) and heterotrophic nitrification aerobic denitrification (HNAD) (An et al., 2020; Yang et al., 2017).

As a new species of bacteria discovered in recent years, the HNAD combines the heterotrophic nitrification (NH$_4^{+}$→NO$_3^{-}$ or NO$_2^{-}$) and simultaneous denitrification (NO$_2^{-}$ or NO$_3^{-}$→N$_2$ or/N$_2$) under aerobic conditions in a single bioreactor (Li et al., 2015). Thus, the route of nitrogen removal process is remarkably cut down and the NH$_4^{+}$-N removal efficiency is much higher than that of conventional biological process due to its rapid proliferation. The increasing species of HNAD have been found and reported, such as Enterobacter cloacae CF-S27 (Padhi et al., 2017), Pseudomonas Stutzeri SDU10 (Chen et al., 2020), and Acinetobacter sp. Y16 (Huang et al., 2013). The eluted wastewater from the weathered crust rare earth tailing possesses the characteristics of high NH$_4^{+}$-N concentration and low pH in which HNAD bacteria survived can be screened out with the highly efficiency for NH$_4^{+}$-N removal (Wang et al., 2016).

The loss of HNAD bacteria in suspended treatment system is another obstacle factor for stable application. And carrier immobilization is an effective method to enhance biodegradation efficiency and reduce loss of microbes (Lü et al., 2019; Yuan et al., 2015). Many materials such as activated carbon, porous glass, alginate and biochar have been applied as the immobilization carrier (Banerjee et al., 2019; Talha et al., 2018). Compared with other carriers, biochar has the advantages of easy modification, nontoxicity, good availability, and low cost. In addition, biochar-based carrier has higher mass transport of oxygen and nutrients to accelerate the growth of bacteria (Zhang et al., 2016). The immobilization of HNAD on the biochar can promote the bacteria concentration in the system and the complex of HNAD and biochar can be reused, showing a promising rapid method for NH$_4^{+}$-N removal. Isolating the efficient HNAD bacteria and immobilizing it on a biochar carrier that can also adsorb NH$_4^{+}$-N is an important feasible strategy, but few researches have been reported.

In this work, a novel HNAD strain named Pseudomonas fulva K3 (P. fulva) with high NH$_4^{+}$-N removal efficiency was isolated from surroundings of the weathered crust rare earth tailing. And the characteristics of nitrogen removal was investigated. The magnesium-modified biochar (MgBC) was prepared as an adsorbent of NH$_4^{+}$-N and a carrier for P. fulva strain via thermal decomposition under oxygen-limited condition. The kinetics and isotherm of NH$_4^{+}$-N adsorption was established. And the mechanism of the intensified effect of MgBC on the P. fulva was proposed by using BET, SEM, and XPS, providing a new strategy for NH$_4^{+}$-N wastewater eluted from weathered crust rare earth ore.

2. Materials and methods

2.1. Preparation of MgBC

The corncobs used as biomass char precursor were collected from Hubei Province of China. The MgBC was prepared by the following steps: Firstly, the corncobs were dried and crushed to 2 mm particle size. Then the corncob was impregnated by 5.0 g/L NaOH solution for 12 h to remove impurities and introduced hydroxyl on the surface of corncob biomass. And the obtained corncob was washed with distilled water several times to neutral, followed by filtering and drying at 70 °C overnight. Secondly, the as-prepared corncob was immersed by 0.3 mol/L magnesium chloride solution for 4 h with stirring. Finally, the magnesium-bearing corncob was dried at 70 °C for 8 h and decomposed at 450 °C for 2 h.
under anaerobic environment to obtain MgBC. The biochar without modification (BC) was prepared under identical conditions without the addition of magnesium.

2.2. Cultivation of *P. fulva* strain

*P. fulva* strain was separated from soil sample around the weathered crust rare earth tailing with ammonia nitrogen pollution, and cultivated using basal medium in 50 mL conical flask. The basal medium components for *P. fulva* enrichment were as follows: 5.0 g/L sodium citrate (Na\(_3\)C\(_6\)H\(_{5}\)O\(_7\)-2H\(_2\)O), 0.47 g/L (NH\(_4\))\(_2\)SO\(_4\), 1.0 g/L K\(_2\)HPO\(_4\), 2.0 g/L NaCl, 0.5 g/L MgSO\(_4\)-7H\(_2\)O, 0.01 g/L MnSO\(_4\), 0.04 g/L FeSO\(_4\)-7H\(_2\)O. The basal medium was autoclaved for 20 min at 121 °C before cultivation. All reagents used in this work were of analytical reagent grade and purchased from Sinopharm Chemical Reagent Co., Ltd., Shanghai, China.

2.3. NH\(_4\)-N removal performance of *P. fulva* strain

The NH\(_4\)-N removal performance was investigated by batches of conical flask experiments. Briefly, the experimental medium was prepared by changing carbon source, the ratio of carbon to nitrogen (C/N), pH, and NH\(_4\)-N concentration on the basis of basal medium. The OD\(_{600}\) value of enriched *P. fulva* solution in logarithmic phase was adjusted to 1.0 with sterile water. And 2.0 % (v/v%) of enriched solution was inoculated into 50 mL medium in a 100 mL conical flask. The conical flask was transferred in a shaker at different rotate speed and temperature. The samples were obtained at intervals to determine OD\(_{600}\) and then centrifuged at 10000 r/min for 10 min. The supernatant was collected to measure the concentrations of NH\(_4\)-N, nitrite nitrogen (NO\(_2\)-N), nitrate nitrogen (NO\(_3\)-N), and total nitrogen (TN). The NH\(_4\)-N biodegradation efficiency of *P. fulva* was obtained by subtracting the amount of gaseous ammonia generated in alkaline condition.

2.4. NH\(_4\)-N adsorption properties of MgBC

The NH\(_4\)-N adsorption experiments were carried out in 100 mL conical flasks containing 50 mL of the as-prepared medium. Then 1.0 % (wt.%) of MgBC powder was added into the conical flask, followed by transferring them to a shaker at 160 r/min at different temperature. After adsorption process, the sample was filtered by 0.22 µm filter membrane. The supernatant was collected to measure the concentrations of NH\(_4\)-N, and the MgBC adsorbed by NH\(_4\)-N was washed with distilled water and then dried for the subsequent characterization.

Equation (1) can be used to calculate the equilibrium adsorption amount (q\(_e\)) of MgBC (Ma et al. 2017a):

\[
q_e = (C_0 - C_e) \cdot \frac{V}{M}
\]

where C\(_0\) (mg/L) and C\(_e\) (mg/L) are the initial and equilibrium adsorption concentration of NH\(_4\)-N, V (L) is the volume of the solution, and M (g) is the mass of MgBC powder.

2.5. Intensified effect of MgBC on *P. fulva* strain for NH\(_4\)-N removal

The intensified effect of MgBC on *P. fulva* strain was investigated by adding both *P. fulva* and MgBC powder in NH\(_4\)-N containing medium. 2.0 % (v/v%) of enriched *P. fulva* solution was inoculated into the medium, and then 1.0 % MgBC powder (wt.%) was added into the conical flask immediately. The samples were taken out periodically to test the concentration of NH\(_4\)-N. The residue after NH\(_4\)-N removal process was washed and dried for the subsequent characterization. All experiments were repeated in triplicate.

2.6. Analytical methods and characterization

The concentrations of NH\(_4\)-N, NO\(_3\)-N and NO\(_2\)-N and TN were measured via Nessler's Reagent, ultraviolet spectrophotometry, 1-naphthylethlyenediamine dihydrochloride and potassium persulfate by using UV-vis spectrophotometer (UV-2600, Shimadzu, Kyoto, Japan), respectively. The absorbance at 600 nm (OD\(_{600}\)) was monitored to indicate the growth of *P. fulva* strain.
The microstructure of *P. fulva* strain and magnesium-modified biochar were observed by scanning electron microscopy (SEM, JSM-7001F, Shimadzu, Kyotocity, Japan). Especially, the *P. fulva* bacteria solution was centrifuged at 10000 r/min for 10 min, and then washed by distilled water three times. The pentanediol (2.5 % by v/v%) was used to immobilize the bacteria at 4 °C for 24 h. Then the *P. fulva* strain was dehydrated in an ascending serious concentration of ethanol (50%, 70%, 90%, and 100% by v/v%) for 10 min each time, followed by the displacement process with isoamyl acetate for 15 min. Lastly, the *P. fulva* strain was freeze-dried for SEM.

The surface area of biochar samples was measured using Brunauer-Emmett-Teller (BET, Micromeritics ASAP 2460, USA) method through N\textsubscript{2} adsorption isotherms. The X-ray photoelectron spectroscopy (XPS, Thermo Fischer ESCALAB 250Xi, USA) measurements were performed to detect the element valence states of MgBC before and after NH\textsubscript{4}+-N adsorption process.

3. Results and discussion
3.1. NH\textsubscript{4}+-N biodegradation characteristics of *P. fulva* strain

3.1.1. Morphology of *P. fulva* strain and growth characteristic

The morphology of *P. fulva* strain was observed by SEM. As shown in Fig. 1(A), the *P. fulva* isolated from soil sample around the weathered crust rare earth tailing was a rod-shaped bacterium with an average length of 1.8 µm. The growth of *P. fulva* strain was exhibited by recording OD\textsubscript{600} values in basal medium during biodegradation process. According the results shown in Fig. 1(B), Slogistic model was used to describe the growth curve of *P. fulva* strain. The equation of Slogistic model could be described as follows (Chen et al., 2016):

\[
y(t) = \frac{a}{1 + b \cdot e^{-kt}}
\]

where a, b, and k are constants of Slogistic model.

The R\textsuperscript{2} of the fitting curve was 0.991, revealing that *P. fulva* strain could be adequately described the growth characteristic of *P. fulva* strain. The OD\textsubscript{600} increased significantly from 0.18 to 1.33 with the increasing time from 4 to 8 h and then remained stable afterward, indicating that the nutrient substances were almost exhausted and *P. fulva* strain reached stationary phase.

Fig. 1. Scanning electron microscopy images (SEM) (A) and growth curve (B) of *P. fulva* strain

3.1.2. The effect of initial pH on the ammonium biodegradation

The effects of initial pH on the OD\textsubscript{600} of *P. fulva* strain and the NH\textsubscript{4}+-N biodegradation efficiency were investigated under the conditions of temperature of 30 °C, rotate speed of 160 r/min, and initial ammonia concentration of 100 mg/L. According to the results shown in Fig. 2, the initial pH is a significant factor during the biodegradation process. The OD\textsubscript{600} increases with the biodegradation time (Fig. 2(A)). The maximum (around 1.33) OD\textsubscript{600} could be reached under the conditions of pH=7.0, 8.0 and 9.0 at the biodegradation time of 8, 9, and 10 h, respectively. The growth of *P. fulva* strain was severely inhibited in strong acidic environment (pH<5.0) and slightly inhibited in alkaline environment (pH>10.0). As shown in Fig. 2(B), the effect of pH on NH\textsubscript{4}+-N biodegradation rate was consistent with that of OD\textsubscript{600}. The NH\textsubscript{4}+-N can be completely removed in neutral and alkaline condition (pH=6.0~10.0),
and the maximum NH$_4^+$-N biodegradation rate could be obtained at pH=7.0. The NH$_4^+$-N is existed as ionic form in strong acidic environment, resulting in the unavailability for ammonia monooxygenase (Amo) in _P. fulva_ strain. As pH increases, more molecular ammonia nitrogen was generated. Therefore, the biodegradation efficiency increases with the increasing pH value. However, the charge of cell membrane can be destroyed by excessive hydroxyl, leading to the decrease of NH$_4^+$-N biodegradation efficiency.

3.1.3. The effect of temperature on the ammonium biodegradation

The structure of bacteria, enzyme activity, substance transferring, and protein synthesis can be greatly impacted by temperature and further influence the NH$_4^+$-N biodegradation. Fig. 3 exhibited the effect of temperature on NH$_4^+$-N biodegradation efficiency under the conditions of pH=7.0, rotate speed of 160 r/min, and initial NH$_4^+$-N concentration of 100 mg/L. The NH$_4^+$-N biodegradation efficiency could be significantly influenced by temperature. The NH$_4^+$-N biodegradation increased with the increase of temperature ranging from 20 to 30 °C due to the improvement of enzyme activity and the diffusivity of substances, and then decreased in the range of 30 to 40 °C because of the denaturation of enzymes and proteins. The time of biodegradation for _P. fulva_ was shortest at the temperature of 30 °C, indicating that the _P. fulva_ strain could grow well for NH$_4^+$-N removal.

3.1.4. The effect of initial NH$_4^+$-N concentration on ammonium biodegradation

The effect of initial NH$_4^+$-N concentration (50~300 mg/L) on ammonium biodegradation was shown in Fig. 4. From the results in Fig. 4(A), the biodegradation time for NH$_4^+$-N removal increased from 7 h to 60 h with the increasing NH$_4^+$-N concentration from 50 mg/L to 300 mg/L. As shown in Fig. 4(B), the maximum removal rate of 34.51 mg/L/h was obtained under the initial NH$_4^+$-N concentration of 150 mg/L, which was much higher than some reported researches, such as _Pseudomonas mendocina_ X49 with a 26.39 mg/L/h maximum rate (Xie et al., 2021), and _Pseudomonas_ sp. ADN-42 with a 11.6 mg/L/h maximum rate (Jin et al., 2015). In addition, the NH$_4^+$-N maximum removal rates firstly increased (initial NH$_4^+$-N concentration=50~150 mg/L) and then decreased (initial NH$_4^+$-N concentration 200~300 mg/L).
mg/L). And the time for corresponding maximum removal rate shifted from 4 h to 12 h, indicating that the initial NH$_4^+$-N concentration higher than 200 mg/L could burden the load of NH$_4^+$-N and inhibit the growth of _P. fulva_ strain.

Fig. 4. The effect of initial NH$_4^+$-N on biodegradation process

### 3.2. NH$_4^+$-N adsorption behaviors of MgBC

#### 3.2.1. Characterization of BC and MgBC

Fig. 5(A) and Fig. 5(B) displayed the SEM images of BC and MgBC, respectively. The morphology of BC changed remarkably after modified by magnesium. The biochar without modification were observed to be composed of small knots with layer structure, and the surface was quite smooth. While the magnesium-modified biochar exhibited a relatively rougher surface due to the pyrolysis decomposition of MgCl$_2$. The N$_2$ adsorption isotherm curve of MgBC measured by BET is shown in Fig. 5(C). The specific surface area of as-prepared MgBC is 6.37 m$^2$/g with an adsorption average pore diameter of 8.50 nm (the inset of Fig. 5(C)), providing a large amount of adsorption sites for NH$_4^+$-N and _P. fulva_ bacteria. The EDS result of the MgBC is shown in Fig. 5(D). The appearance of Mg element reveals that the biochar is successfully modified by Mg.

Fig. 5. The SEM images of BC(A), and MgBC(B), N$_2$ adsorption isotherm curve of MgBC (C), EDS of MgBC (D)
3.2.2. NH$_4^+$-N adsorption kinetics and isotherms

Two kinetic models, namely Lagergren’s pseudo-first-order model (Eq. 3) and pseudo-second-order kinetic model (Eq. 4), were used to study the adsorption behaviors of MgBC (Li et al. 2019). And the equations are expressed as follows:

\[
\ln(q_e - q_t) = -K_1t + \ln q_t \\
\frac{1}{q_t} = \frac{1}{K_2q_e^2t} + \frac{1}{q_e}
\]

where \(q_t\) (mg g\(^{-1}\)) and \(q_e\) (mg g\(^{-1}\)) are the amount of the adsorbed NH$_4^+$-N at time \(t\) (min) and at equilibrium, \(K_1\) (min\(^{-1}\)) and \(K_2\) (min\(^{-2}\)) are the adsorption rate constant of the pseudo-first-order equation and pseudo-second-order equation.

The NH$_4^+$-N adsorption kinetics were carried out in basal medium under the conditions of initial NH$_4^+$-N concentration of 100 mg/L, temperature of 30 °C, and the MgBC dosage of 1.0 % (wt%). The equilibrium was achieved within 180 min with a maximum adsorption capacity of 2.23 mg/g. The adsorption kinetics models fitted by pseudo-first-order model and pseudo-second-order model were exhibited in Fig. 6(A) and Fig. 6(B), respectively. And the kinetics parameters were listed in Table 1. The pseudo-second-order model fitted the experimental data better due to the higher correlation coefficient \((R^2)\). And the value of \(q_e\) (2.47 mg g\(^{-1}\)) calculated by the pseudo-second-order is also more approximate to the experimental value (2.23 mg g\(^{-1}\)), which indicates that the NH$_4^+$-N adsorption process is mainly controlled by chemical adsorption (Wang et al., 2018).

\[
\begin{align*}
\text{C}e & = \frac{1}{K_L} q_{\text{max}} c_e + \frac{1}{q_{\text{max}}} \\
q_e & = \frac{1}{K_L} c_e + \frac{1}{q_{\text{max}}} \\
\end{align*}
\]

The adsorption isotherms were fitted by Langmuir (Eq. (5)) and Freundlich (Eq. (6)) models, which were presented as follows:

\[
\begin{align*}
\frac{1}{q_e} &= K_L c_e + \frac{1}{q_{\text{max}}} \\
\ln q_e &= \ln q_{\text{max}} + \frac{1}{K_L} \ln c_e
\end{align*}
\]

where \(C_e\) (mg L\(^{-1}\)) is the equilibrium concentration of NH$_4^+$-N in medium, \(q_e\) (mg g\(^{-1}\)) is the amount of NH$_4^+$-N adsorbed on the MgBC, \(q_{\text{max}}\) (mg g\(^{-1}\)) is the maximum adsorption amount and \(K_L\) (L/mg) is a constant related to Langmuir model.

Fig. 6. Pseudo-first-order (A) and pseudo-second-order (B) kinetics, Langmuir isotherms (C) and Freundlich isotherms (D) for NH$_4^+$-N adsorption on MgBC
\[
\ln q_e = \ln K_f + \frac{\ln c_0}{n}
\]

where \(K_f\) (mg\((1-1/n)\)g\(-1\)L\(^{-1}\)/n) is the Freundlich constant related to the adsorption capacity and \(n\) is a constant which feature the system.

The adsorption isotherms fitted by Langmuir and Freundlich methods were displayed in Fig. 6(C) and Fig. 6(D), respectively. And the adsorption isotherms parameters were listed in Table 2. The correlation coefficient obtained by Freundlich fitting was 0.995, higher than that of Langmuir model (0.951), indicating that Freundlich isotherm fitted better than Langmuir isotherm. Therefore, the NH\(_4\)\(^+\)-N adsorption on MgBC is determined to be a homogenous and multilayer adsorption process. In addition, the value of \(n\) in Freundlich equation is calculated to be 0.197, which is in the range of 0.1-0.5, suggesting that the NH\(_4\)\(^+\)-N can be easily adsorbed by MgBC.

### 3.2.3. Mechanism of NH\(_4\)\(^+\)-N adsorption

XPS analysis was conducted to further investigate the mechanism of NH\(_4\)\(^+\)-N adsorption on MgBC. Fig. 7(A) showed the survey spectra of MgBC before and after NH\(_4\)\(^+\)-N adsorption. The main elements on the surface of MgBC were C, O, Mg, and Cl, which were consistent with the results obtained by EDS (Fig. 5(D)). The XPS intensities of Mg, O, and Cl were all diminished after adsorption process due to the dissolution of undecomposed MgCl\(_2\). And N element could be observed after adsorption process, indicating that NH\(_4\)\(^+\)-N was successfully adsorbed on the surface. Fig. 7(B), Fig. 7(C), Fig. 7(D), Fig. 7(E) and Fig. 7(F) exhibited the high solution spectra of C\(_1\)s, O\(_1\)s, Mg\(_{1}\)s, P\(_{2}\)p and N\(_{1}\)s, respectively. The chemical environment of C (Fig. 7(B)) almost keep unchanged, revealing that the NH\(_4\)\(^+\)-N could not be adsorbed on C of MgBC by chemical adsorption. According to the results from Fig. 7(C), the peaks at 532.9 eV and 530.6 eV were ascribed to C\(_{\text{O}}\) and Mg\(_{\text{O}}\) of MgBC and then disappeared after the adsorption process. The binding energy at 533.9 eV and 531.8 eV were ascribed to O\(_{\text{O}}\)-NH\(_4\) adsorbed on MgBC, indicating that the PO\(_3\)\(^{-}\) in basel medium can work as a “bridge” to further adsorb NH\(_4\)\(^+\)-N. Fig. 7(D) showed the high solution XPS spectra of Mg\(_{\text{I}}\)s. The peak at 1305.3 eV corresponded to Mg\(_{\text{O}}\), suggesting that Mg was existed as Mg\(_{\text{O}}\) before adsorption process. After the adsorption process, the peak Mg\(_{\text{O}}\)-NH\(_4\)(1306.0 eV) and Mg\(_{\text{O}}\)-P(1304.7 eV) were observed, indicating that NH\(_4\)\(^+\)-N and PO\(_3\)\(^{-}\) could be adsorbed on MgBC. In addition, Fig. 7(E) and Fig. 7(F) can further confirm the adsorption of N and P on the surface of MgBC by comparison of the intensity of XPS spectra before and after adsorption process.

### 3.3. NH\(_4\)\(^+\)-N removal characteristics of eluted water from weathered crust rare earth tailing

The eluted water was collected from weathered crust rare earth tailing with the NH\(_4\)\(^+\)-N concentration of 94 mg/L. Fig. 8(A) exhibited the NH\(_4\)\(^+\)-N removal efficiency after 5 h biodegradation under different pH ranging from 5.0 to 11.0 using MgBC (the blue), \(\textit{P. fulva}\) (the red) and MgBC + \(\textit{P. fulva}\) (the black), respectively. The NH\(_4\)\(^+\)-N removal efficiency increased with the increasing pH in the range of 5~10 due to the elimination of competitive adsorption of H\(^+\). And then decreased because of the existing form of NH\(_3\)-H\(_2\)O when the pH higher than 10. The optimum pH was 7.0 for \(\textit{P. fulva}\) strain, which was in accordance with the results obtained in Fig. 2. The optimum pH for MgBC+\(\textit{P. fulva}\) was chosen to be 7.0~8.0. As shown in Fig. 8(B), the NH\(_4\)\(^+\)-N removal efficiency firstly increased to 21.6 % due to the chemical adsorption on MgBC in 3 h, and then removed via synergetic effect of \(\textit{P. fulva}\) biodegradation and MgBC adsorption in the following time. And the consuming time for NH\(_4\)\(^+\)-N removal was shorten to be 7 h due to the synergetic effect of MgBC and \(\textit{P. fulva}\) strain. The nitrogen transformation characteristics in nitrogen removal process was shown in Fig. 8 (C). The NH\(_4\)\(^+\)-N concentration decreased from 94.2 to 3.11 mg/L in 7.0 h, and NO\(_3\)-N was generated at 5~6 h due to the heterotrophic nitrification of \(\textit{P. fulva}\) strain. NO\(_3\)-N was detected lower than 0.3 mg/L in the whole process. At 10.0 h, TN reached to the minimum concentration of 14.4 mg/L with a removal efficiency of 84.7 %. The SEM (Fig. 8 (D)) was carried out to investigated the microstructure of MgBC+\(\textit{P. fulva}\). The NH\(_4\)\(^+\)-N could be adsorbed on the surface of MgBC, and \(\textit{P. fulva}\) was also attached to the surface of MgBC, consuming the NH\(_4\)\(^+\)-N adsorbed on MgBC and breaking the adsorption equilibrium. Thus, more NH\(_4\)\(^+\)-N could be adsorbed on MgBC. The intensified effect for NH\(_4\)\(^+\)-N removal was achieved by \(\textit{P. fulva}\) and MgBC.
Fig. 7. XPS spectra of MgBC before and after adsorption; survey spectra(A); high solution spectra of C1s(B), O1s(C), Mg1s(D), P2p(E), and N1s(F)

Fig. 8. The effect of pH (A) and time (B) on NH₄⁺-N removal using MgBC, *P. fulva* and MgBC+*P. fulva*, the nitrogen transformation characteristics (C), and microstructure of MgBC+*P. fulva* after NH₄⁺-N removal process.
3.4. Mechanism of the intensified effect for NH\textsubscript{4}\textsuperscript{+}-N removal

According to the XPS and SEM results, intensified removal mechanism between MgBC and *P. fulva* was proposed in Fig. 9 (A). The MgBC provides a large amount of specific surface area for NH\textsubscript{4}\textsuperscript{+}-N adsorption and attachment sites for *P. fulva*. The NH\textsubscript{4}\textsuperscript{+} and PO\textsubscript{4}\textsuperscript{3-} are adsorbed on the O and Mg, respectively. And NH\textsubscript{4}\textsuperscript{+} can be further attached to the O atom of PO\textsubscript{4}\textsuperscript{3-}, which works as a “bridge” to adsorb more NH\textsubscript{4}\textsuperscript{+} on MgBC. The NH\textsubscript{4}\textsuperscript{+} can be immobilized on MgBC when the initial concentration is high and then released to system when its concentration gets lower due to the consuming of *P. fulva*, keeping a proper NH\textsubscript{4}\textsuperscript{+} concentration to facilitate the growth of *P. fulva* strain. The intensified effect for nitrogen removal can be achieved by the cooperation between MgBC and *P. fulva* strain.

![Synergistic removal mechanism of MgBC in the present of P. fulva strain (A), schematic diagram of the electron transport chain of P. fulva](image)

Fig. 9. Synergistic removal mechanism of MgBC in the present of *P. fulva* strain (A), schematic diagram of the electron transport chain of *P. fulva*

The electron transport of chain of *P. fulva* is exhibited in Fig. 9 (B) according to the nitrogen transformation characteristics in experiments. Firstly, NH\textsubscript{4}\textsuperscript{+}-N is oxidized to NH\textsubscript{2}OH in the presence of ammonia single oxidase (AMO), and NH\textsubscript{2}OH can be further oxidized to NO\textsubscript{2}\textsuperscript{-} and NO\textsubscript{3}\textsuperscript{-} with the help of hydroxylamine oxidase (HAO). Then aerobic denitrification process occurs, reducing NO\textsubscript{2}\textsuperscript{-} and NO\textsubscript{3}\textsuperscript{-} to N\textsubscript{2} under serious enzymes such as nitrite reductase (NIR), nitrate reductase (NAR), nitric oxide reductase (NOR) and nitric oxide reductase (NOS). The NH\textsubscript{4}\textsuperscript{+}-N in eluted water from weathered crust rare earth tailing is successfully removed by the intensified effect of MgBC and *P. fulva* strain with great efficiency.

4. Conclusions

In summary, the isolated strain *P. fulva* and the as-prepared MgBC both have the ability of NH\textsubscript{4}\textsuperscript{+}-N removal. The effects of pH, temperature, initial NH\textsubscript{4}\textsuperscript{+}-N concentration on *P. fulva* biodegradation process were investigated, and the adsorption properties of MgBC was studied. The mechanism of the intensified effect of MgBC on *P. fulva* was revealed using XPS, BET, and SEM.

(1) The isolated *P. fulva* exhibited a rod-shaped morphology with an average length of 1.8 μm. NH\textsubscript{4}\textsuperscript{+}-N can be completely biodegraded under the optimal conditions of pH=7.0~8.0, temperature 30 °C, and initial NH\textsubscript{4}\textsuperscript{+}-N concentration of 100~150 mg/L. The NH\textsubscript{4}\textsuperscript{+} tolerant concentration for *P.
fulva was determined to be 300 mg/L, showing great feasibility for the treatment of eluted water from weathered crust rare earth tailing.

(2) The NH$_4^+$-N adsorption properties of MgBC were studied. Pseudo-second-order model is more suitable to describe this adsorption kinetic with a higher correlation coefficient, and the Freundlich model fits the isotherm data well in adsorption process, implying that the adsorption of is a multilayer adsorption process mainly controlled by chemical adsorption. The mechanism of NH$_4^+$-N adsorption was confirmed by XPS results. NH$_4^+$-N was adsorbed to the O element of MgBC, and the PO$_4^{3-}$ adsorbed on Mg element may act as a “bridge” to further adsorb the NH$_4^+$-N.

(3) The mechanism of the intensified effect between MgBC and P. fulva strain was revealed. The MgBC provides a large amount of specific surface area for NH$_4^+$-N adsorption and attachment sites for P. fulva. The NH$_4^+$-N can be immobilized on MgBC when the initial concentration is high and then released to system when its concentration gets lower due to the consuming of P. fulva, keeping a proper NH$_4^+$-N concentration to facilitate the growth of P. fulva strain. The total nitrogen removal efficiency of the eluted weathered crust rare earth tailing reached 84.7 % using the complex of MgBC and P. fulva.

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