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OBȚINEREA BIO-NANOPARTICULELOR DE FOSFAȚI DE MAGNEZIU ȘI PROPRIETĂȚILE LOR LIANTE
PREPARATION OF BIOLOGICAL MAGNESIUM PHOSPHATES NANOPARTICLES AND THEIR BINDING PROPERTIES

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Magnesium phosphates can be prepared by microbial induced deposition, and bio-grouting and stirring processes are used to bind loose sand particles. The result shows that loose sand particles can well be cemented by stirring method. However, loose particles are not bound by pumping method. The X-ray diffraction (XRD) of the precipitated slurry indicates that the main component is a mixture of hydrated magnesium phosphate. Energy dispersive spectrometer (EDS) displays that elements of sandstones are mainly C, Si, O, P, Mg, Na and Cl. The average compressive strength and porosity of sandstones are 0.99±0.3MPa and 26.3±0.5%, respectively. Scanning electron microscope (SEM) shows that the cementation material in sandstones is flake nano-clusters with the size in the range of 0.5-1.5μm. Transmission electron microscopy (TEM) further indicates that a single nanoparticle is like irregular sheet in morphology, and the size of the cementitious material is in the range of 200-500 nm.

Keywords: *magnesium phosphates; pumping method; stirring method; sandstone; compressive strength; Transmission Electron Microscopy*

1. Introduction

Portland cement and bio-cement are important cementitious materials in the construction sector [1-7]. The carbon dioxide and ammonia is released in the production process of Portland cement and bio-carbonate cement, respectively [8-10]. A large quantity of carbon dioxide is emitted in the production process of cement and can cause greenhouse effect. 2 mol of ammonia are produced and released into the air when 1mol of calcium carbonate is prepared by microbial induced precipitation. These ammonia gas has a negative impact on our human health and the environment. Therefore, a new cementitious material should be selected to replace partially Portland cement and bio-carbonate cement. Finally, bio-phosphate cement (microbial-induced deposition phosphate) is selected because of the presence of phosphate minerals in nature.

In the paper, biological magnesium phosphates were prepared by *Bacillus subtilis* inducing precipitation. The biological magnesium phosphate was applied to bind loose sand particles into a bio-sandstone by bio-grouting process and stirring process, and the bio-sandstones have a certain mechanical properties. The structure and morphology of the biological magnesium phosphate was systematically studied by X-ray technique (XRD), scanning electron microscope (SEM), and transmission electron microscopy (TEM).

2. Experimental

2.1. Materials

All raw materials were purchased without further purification. Deionized water was self-made. *Bacillus subtilis* was purchased from China Center of Industrial Culture Collection. Phosphate monoester, beef extract, peptone and sodium chloride were purchased from Sinopharm Chemical Reagent Co., Ltd. The aggregate used in this study was quartz sand whose particle radius is less than 300μm (grain size characteristics: d_{10}= 150μm (10% of the grains were lower than a diameter of this size); d_{90}= 300μm).

2.2. Preparation of magnesium phosphates

Microbial-induced precipitation of magnesium phosphates: *Bacillus subtilis* was cultivated by a medium containing 3g/L of beef extract, 5g/L of peptone, and 3g/L of sodium chloride. Finally, the harvested microorganisms were kept in a refrigerator at 4°C for stock prior to use.

20mM of phosphate monoester was dissolved in 100ml of bacterial solution, and it was

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allowed to stand under static conditions for 2 hours at 30±2°C in an incubator. $MgCl_2$ (10mM) was added to the above solution. The precipitates were allowed to stand under static conditions for 24 hours at room temperature. The products were filtrated and dried at 60±2 °C for 24 h in an oven. As a result, 0.73g of magnesium phosphates was obtained.

2.3. Specimens preparation

Magnesium phosphates, which the quality accounted for 40% of the total mass of quartz sand, was mixed with quartz sand, and mechanical stirring until completely homogeneous [9]. Mixed sand mortar was then poured into 60ml plastic syringe (Φ3cm×6cm), as is shown in Figure 1. The specimens were kept in an oven at 60±2°C for 14days. Afterwards, the bio-sandstones were obtained after demold (Fig. 2).

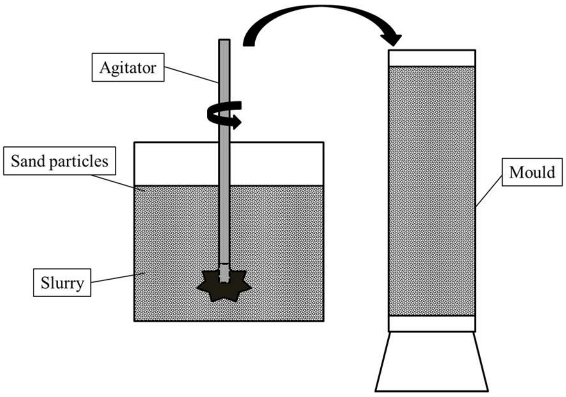


Fig. 1 - Schematic diagram of mixing process.

Fig. 2 - Images of the bio-sandstones obtained by mixing process.

25 ml of the mixed solution of phosphate monoester and bacteria was firstly injected in sand column (Φ3cm×6cm). Next, 25 ml of $MgCl_2$ (0.1mol/l) was injected into the sand column. After standing 6 h, 25 ml of the mixed solution was injected to sand column, and then 25 ml of $MgCl_2$ (0.1mol/l) was injected, as is shown in Figure 3. Above steps were repeated for 22 times. The specimens were kept in an oven at 60±2°C for 14d. Afterwards, the bio-sandstones were obtained after demold (Fig. 4)

Fig. 3 - Sand column cemented by bio-grouting process.

Fig. 4 - Images of the bio-sandstones obtained by bio-grouting process.

2.4. Characterization

X-ray techniques (XRD, Bruker Company, Germany) analysis was carried out on sample at room temperature by a D8-Discover X diffractometer (40kv, 40mA) with Cu (λ=1.5406Å) irradiation at the rate of 0.15s/step in the range of 5-90˚. Scanning electron microscope (SEM, FEI Company, Netherlands, operating voltage 20 kV) with a Genesis 60S energy dispersive X-ray spectroscopy (EDS) spectroscopy system was used to conduct morphological studies and to measure the elemental compositions of the samples. Transmission electron microscopy (TEM) images were obtained on a FEI, G2 20 equipment. TEM grids were prepared using a few drops of nanoparticles followed by drying. The compressive strength of bio-sandstone was tested via computer-controlled electronic universal testing machine (SANS CMT 8502) with a loading rate of 1 mm/min. The average porosity of bio-sandstones was estimated using the formula [11, 12],

$$p = \frac{M_{sat} - M_{dry}}{\rho_w V_{vol}}$$

Where P is the porosity of bio-sandstones, M_{sat} is the weight of the bio-sandstones as it was fully saturated with water (g), M_{dry} is the weight of

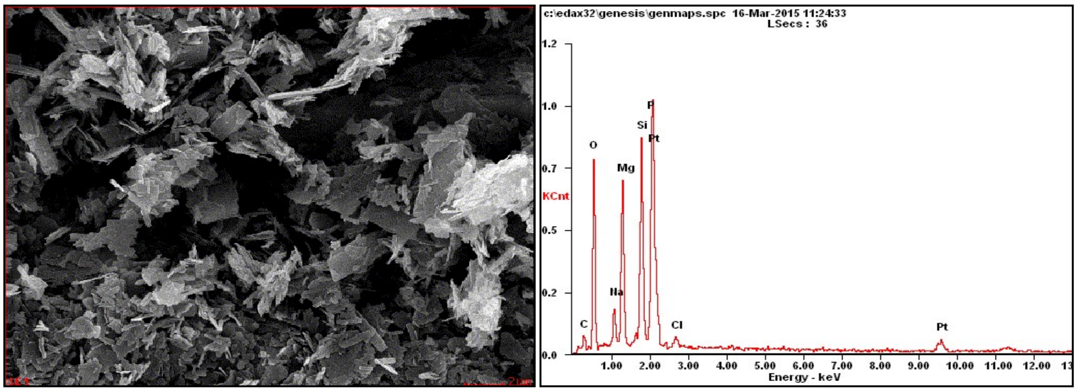


Fig. 5 - SEM image and EDS spectrum of the specimen.

the dried bio-sandstones (g), V_{vol} is the volume of bio-sandstones (cm^3), ρ_w is the density of water (1.0 g/cm^3).

3. Results and discussions

Magnesium phosphates nanoparticles can be prepared by *Bacillus subtilis* inducing precipitation. The formation mechanism of magnesium phosphates can be explained by the following steps: (1) Phosphate monoester is hydrolyzed by alkaline phosphatase secreted by *Bacillus subtilis*, and then phosphate ions are obtained; (2) magnesium phosphates are forming by phosphate ions reaction with magnesium ions.

An elemental analysis of the specimens' composition was performed by EDS spectrum to confirm the presence of elements C, O, Na, Si, P, Cl and Mg in the samples, as shown in Figure 5. The XRD analyze of the sample confirms that the cementation material is the mixture of hydrated magnesium phosphate, such are: $Mg_3(PO_4)_2{\cdot}8H_2O$, $Mg_3(PO_4)_2{\cdot}22H_2O$ and $Mg_3(PO_4)_2{\cdot}5H_2O$, corresponding to JCPDS Card No. 16-0330, 31-0805, and 05-0628, respectively (Fig. 6). The impurity was sodium chloride NaCl (JCPDS Card No. 05-0628).

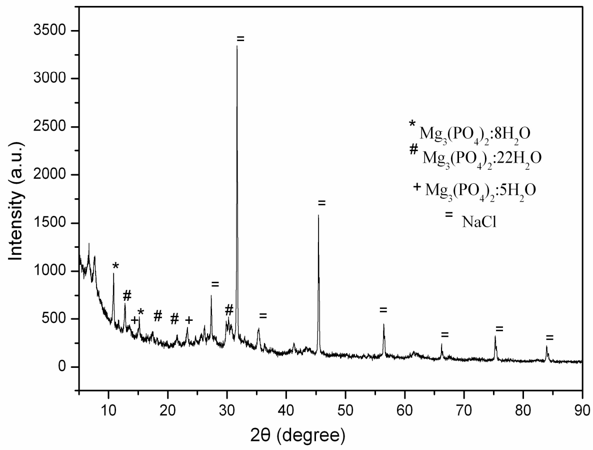
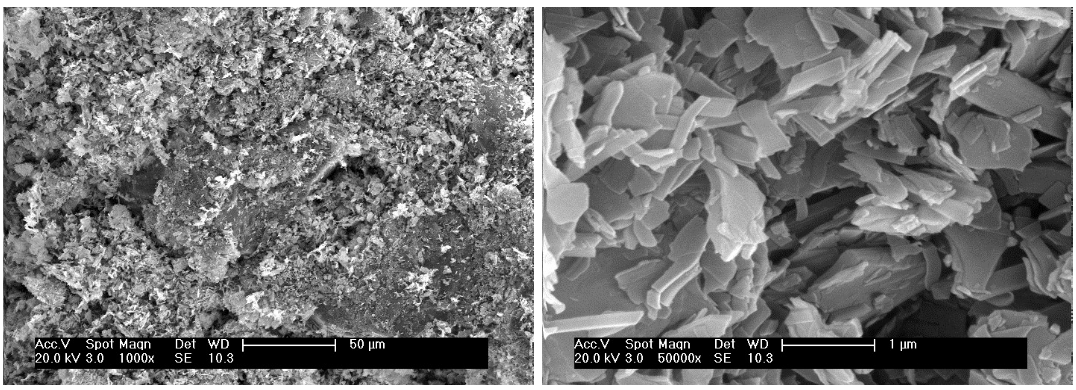


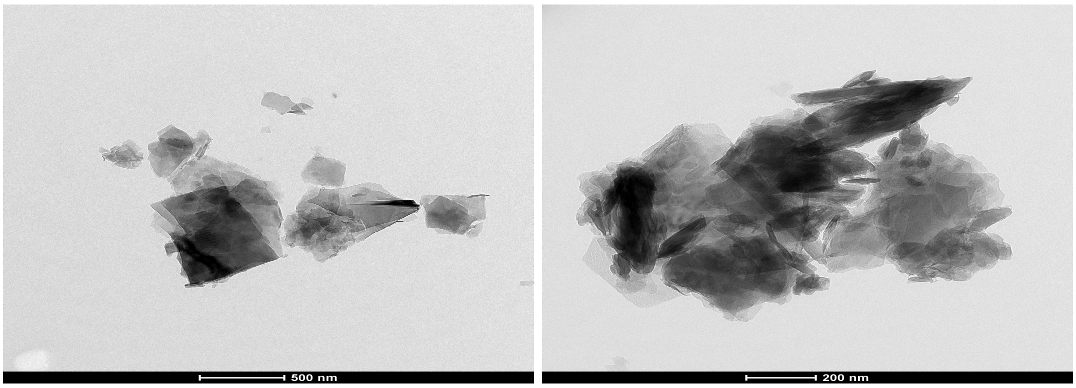
Fig. 6. - XRD pattern of the sample.

Loose sand particles can be cemented by mixing process and bio-grouting process. Each group of specimen including three bio-sandstones was used to determine the compressive strength. The average compressive strength of two types of bio-sandstones cemented by mixing process and bio-grouting process is 0.99±0.3MPa and 0.06±0.02MPa, respectively. The strength of two



(a) (b)

Fig. 7 continues onnext page



(c) (d)

Fig. 7 -SEM images (a and b) and TEM images (c and d) of the bio-sandstones cemented by mixing process.

types of bio-sandstones is lower than those of the regular magnesium phosphate cement [13]. Porosity of two types of bio-sandstones is effectively reduced from 39.8±0.5% and 40.1±0.7% down to 26.3±0.5% and 38.7±0.7% respectively. Flocculent sediments can be filled out in bio-grouting process. Therefore, the strength of the bio-sandstones cemented by bio-grouting process is lower than those cemented by mixing process.

SEM and TEM images of the bio-sandstones cemented by mixing process are shown in Figure 7. The morphology of the bio-sandstones is characterized by flake particles with size in the range of 0.5-1μm, as is shown in Figures 7a and b. TEM images further display that magnesium phosphates nanoparticles are mainly irregular flakes in shape with sizes in the range of 200-500nm (Figs. 7c and 7d).

4. Conclusions

Loose sand particles can be cemented into bio-sandstone by nano-magnesium phosphates. The morphology of nano-magnesium phosphates is characterized by irregular flakes in shape with sizes in the range of 200-500nm. The strength of the bio-sandstones cemented by bio-grouting process is lower than those cemented by mixing process. The average compressive strength and porosity of the bio-sandstones cemented by mixing process is 0.99±0.3 MPa and 26.3±0.5%, respectively. Along with the maturation of the method in future studies, there will be multiple new opportunities for engineering applications, for instance, desert control, bluff, slope, soil stabilization, and so on.

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