Self-healing technology based on microbial induced carbonate precipitation can achieve cracks-healing of concrete. As concrete cracks appear, dormant bacterial spores introduced into concrete are activated, and calcium lactate is used as substrate to form calcium carbonate for healing crack. Due to the harsh environment in concrete, bacterial spores directly introduced become inactivated. Therefore, the introduction of good protective carrier is critical, and the mechanical properties and economy of the carriers are also the keys to improving self-healing technology. In this study, recycled aggregate was used as a protective carrier for Bacillus pasteurii to enhance the self-healing capacity. The effects of this technique were investigated by comparing with three other incorporation techniques, i.e., direct introduction of bacteria, diatomaceous earth-immobilized bacteria, and expanded perlite-immobilized bacteria. The healed crack width value of specimens incorporated with recycled aggregate-immobilized bacteria was close to that of specimens incorporated with expanded perlite-immobilized bacteria (the healed widths were 0.28 mm and 0.32 mm, respectively), which was larger than that of specimens incorporated with diatomaceous earth-immobilized bacteria (the healed width was 0.14 mm) and specimens directly introduced bacteria. Scanning electron microscope and electronic data switching analysis confirmed that precipitation formed at cracks was calcium carbonate.

1. Introduction

In recent years, with the rapid development of the construction industry, concrete has been widely used due to its high compressive strength and low price compared with other construction materials. However, with low tensile strength (Mehta 2008; Yazdanbakhsh et al. 2016; Khaliq et al. 2016; Nie et al. 2019; Wang et al. 2019), it may cause micro-cracks easily during the service life, which will hamper durability of concrete structures. Further, cracked concrete is more susceptible to penetration of some corrosive ions, such as chloride and sulfate (Wang et al. 2020). The intrusion of these harmful ions directly leads to concrete deterioration and steel reinforcement corrosion. Therefore, crack healing has become the focus of researchers. Most of current repair methods applied are divided into autogenous healing and manual repairing. However, traditional manual repairing methods, such as manual grouting, surface treatment, and filling do not diagnose and repair cracks in time, and these methods are suitable for repairing larger cracks rather than micro cracks (Yu 2016). Besides, the cracks repaired with the above methods may crack repeatedly over time.

Some studies reported that some small cracks in concrete can heal under certain circumstances (Ahn et al. 2010; Mihashi et al. 2012; Wu et al. 2012; Luthfi et al. 2018; Xu et al. 2018; Zdeb et al. 2018). The reason for this phenomenon was that the crack was filled with calcium carbonate formed by self-healing technology. Self-healing concrete is based on the development of microbial induced carbonate precipitation (MICP) technology (Wang et al. 2017). Some scholars have tried to use some bacteria’s respiration to induce the production of calcium carbonate crystals to repair concrete cracks. As a common microorganism of MICP, Bacillus pasteurii can maintain strong activity under harsh environmental conditions such as strong alkali and high salinity. Once concrete cracks appear, the dormant bacterial spores immobilized in the carriers restore metabolic function, using calcium lactate as substrate to form calcium carbonate. The formation process of CaCO₃ can be described as follows:

\[ \text{Substrate} + \text{Calcium lactate} + \text{O}_2 \xrightarrow{\text{Bacteria}} \text{CaCO}_3 + \text{CO}_2 + \text{H}_2\text{O} \]  \hspace{1cm} (1)

Based on the above principle, some specific self-healing agents were introduced into the matrix to explore healing capacity. To date, some bacteria have been widely used as healing agents to enhance the healing of cracks (Chen et al. 2016). Wang et al. (2016) applied bacteria-induced CaCO₃ precipitation for concrete surface protection and cracks repair, revealing that biological CaCO₃ as a surface coating and crack sealing
material can greatly reduce permeability to improve the durability of concrete. The microbial induced carbonate precipitation technology could independently diagnose the formation of cracks and achieve self-healing of cracks in concrete structures without delay (Aïssa et al. 2012; Han et al. 2015; Qian et al. 2015; Lee et al. 2018; Li et al. 2018; Li et al. 2020). Jonkers et al. (2010) firstly incorporated bacterial spores and calcium lactate directly into concrete. The results showed that incorporated bacteria and calcium lactate did not negatively affect concrete compressive strength. However, the authors also observed that microbial survival rate dropped by more than 90% within one month. Bacterial spores became crushed or inactivated due to high alkalinity, leading to decrease of viability and mineralization capacity. Subsequently, they encapsulated Bacillus and calcium lactate in ceramsite and then added it to concrete. The results showed that ceramsite prolonged the survival life of Bacillus in concrete and significantly improved the self-healing capacity of concrete. Zhang et al. (2017) developed a self-healing concrete with bacteria immobilized in porous expanded perlite particles. After 28 days of incubation, the value of completely healed crack width was up to 0.79 mm, which was larger than the value of 0.39 mm for specimens that were directly introduced bacteria without protective carrier. The above research results showed that introduction of the carrier provided a porous and low alkaline environment for the growth of Bacillus, and improved microbial induced carbonate precipitation. Therefore, to develop self-healing technology and reduce structural cracking, scholars are actively exploring the self-repairing agent carrier that is more conducive to mineral-forming capacity (Li and Liu 2017; Li et al. 2017; Alazharia et al. 2018; He et al. 2020).

Various protective carriers, such as ceramsite (Xu et al. 2018), expanded clay particles (Amiri et al. 2018), expanded perlite (EP) particles (Qian et al. 2017; Wiktor et al. 2017; Zhang et al. 2017), and diatomaceous earth (DE) (Wang et al. 2012, 2014) have been proposed for enhancing the self-healing capacity of concrete. However, the mechanical properties of protective carriers are lower, leading to limitation of microbial concrete application. On the other hand, the difference of carrier nature results in low compatibility with concrete (Li et al. 2018). Therefore, it is necessary to explore more practical carriers for enhancing the carbonate precipitation of self-repairing concrete.

Recycled aggregate (RA) is a product derived from the condensation of natural aggregates and cement-based hydrates in abandoned buildings (Xiao et al. 2018). RA with hardened cement mortar has higher porosity and water absorption than natural aggregates (Shi et al. 2016). A large amount of old mortar attached to the surface of RA was carbonized for a long time, which caused the surface alkalinity of RA to decrease. Actually, this pore space and low alkaline environment are more conducive to the survival of microorganisms. The effectiveness of this technique was investigated by comparing with three other incorporation techniques, i.e., direct introduction of bacteria, diatomaceous earth-immobilized bacteria (DE-immobilized bacteria) and expanded perlite-immobilized bacteria (EP-immobilized bacteria). In addition, the mechanical properties of the RA are better than those of the carriers used by the other researchers (Butler et al. 2017; Chakradhara et al. 2017; Edvardsen et al. 2017; Liu et al. 2019a, 2019b, 2019c). Based on the above considerations, the use of RA as a protective bacterial carrier to achieve crack self-healing will promote the development of self-repairing concrete.

2. Materials and methods

2.1 Self-healing agent preparation

*Bacillus pasteurii* (A484) was used in this study. Bacterial strains were first rejuvenated by the agar streak method and then cultured in a liquid medium consisting of 5 g peptone, 3 g beef extract, 0.42 g NaHCO₃, and 0.53 g NaCO₃ per liter of distilled water. The medium was sterilized by autoclaving for 30 min at 121°C, and the pH was adjusted to 8.5. Cultures were aerobically incubated at 30°C in a water bath shaker at 120 rpm for 24 h. Spore formation was confirmed by phase microscopy. The OD600 value of bacterial suspension reached up to 0.8 (Achal et al. 2013). The concentration of bacterial spores was microscopically measured as 2.8 × 10⁷ cells/ml.

2.2 Preparation of protective carriers

Three kinds of protective carriers were all obtained from Xi’an, China. The properties of the three carriers were shown in Table 1. In this study, neither recycled aggregate nor expanded perlite was treated. Diatomaceous earth (DE) is a soft natural siliceous sedimentation. In order to maintain the same particle size as other carriers, the diatomaceous earth was treated. After the DE was diluted with water (1:1, v/v), it was dried in the oven at 100°C until a constant weight, and then broken. Particle size of the above three kinds of carriers was all 5 to 8 mm. Impregnation of the particles was carried out under vacuum saturation with the prepared bacterial spore suspension. Vacuum pump was adjusted to a negative pressure of 0.6 MPa for 30 minutes, and then the mixture was removed. Next, all protective carriers were dried in the oven at 45°C for 24 to 36 hours until a constant weight was achieved.

<table>
<thead>
<tr>
<th>Table 1 Properties of carriers.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Carrier types</strong></td>
</tr>
<tr>
<td><strong>Apparent density (kg/m³)</strong></td>
</tr>
<tr>
<td><strong>Water absorption (%)</strong></td>
</tr>
<tr>
<td><strong>Specific surface area (m²/g)</strong></td>
</tr>
<tr>
<td><strong>Pore diameter (µm)</strong></td>
</tr>
<tr>
<td><strong>Porosity (%)</strong></td>
</tr>
<tr>
<td><strong>Numerical tube pressure (MPa)</strong></td>
</tr>
<tr>
<td><strong>Crushing index</strong></td>
</tr>
</tbody>
</table>
2.3 Mix proportions and preparation of specimens

Concrete specimens with dimensions of 40 mm × 40 mm × 160 mm were prepared with ordinary Portland cement (P.O. 42.5, China), coarse aggregate, natural sand and calcium lactate. Recycled coarse aggregate was used as a carrier to immobilize microorganism because of the porosity characteristics of the old mortar and good mechanical strength. Within a certain range, the smaller the carriers particle size, the larger the specific surface area, which was more conducive to the survival of microorganisms. Therefore, the carrier of small particle size was used in this study, and this determined the dimensions of the specimens. In fact, the specimen dimensions of 40 mm × 40 mm × 160 mm was really too small for concrete with coarse aggregate, and it was generally more suitable for mortar specimen without coarse aggregate. Because the apparent healing capacity was not affected by the dimensions of the concrete specimens, the concrete specimens of small-dimensions were used in the early stage, which also saved test costs. These specimens were so light that crack width was favorably controlled, avoiding the change of cracks due to a large dead weight. The purpose is to provide the basis for the research of conventional dimensions concrete specimens and structures. In addition, the use of mortar specimens does not meet the purpose of this study. The cracking mechanism of concrete specimens with coarse aggregates and mortar specimens is different. The application of coarse aggregates lay the foundation for the future crack formation and heal mechanism of concrete specimens, which more truly reflects the cracking and heal mechanism.

The sand was obtained from the Sand and Stone Factory in Xi’an, Shaanxi Province of China. Its fineness modulus and bulk density were 2.6 and 1300 to 1600 kg/m³, respectively. The medium components such as peptone and beef extract were obtained from Microbiology Institute of Shaanxi. The medium was prepared with distilled water and the concrete specimens were prepared with tap water. The water to cement ratio and the water to bacterial suspension ratio of these mixes were 0.48 and 2.4, respectively. The carrier volume was 15% of that of specimens. After 24 h of curing, all the specimens were carefully unmolded, and kept at conditions of 22 ± 2°C and 85 ± 5% humidity for further curing.

Specimens that were directly introduced bacteria into concrete mixture without any protective carrier were named D-C. Test specimens in which bacteria were indirectly introduced into the concrete mixture using RA, EP, and DE as protective carriers were called as R-C, E-C, and B-C, respectively, and the control specimens without bacterial spores were called N-C. Five different types of concrete mixes were prepared, as shown in Table 2.

### Table 2 Mixing proportion of concrete specimens.

<table>
<thead>
<tr>
<th>Specimens numbering</th>
<th>N-C</th>
<th>D-C</th>
<th>E-C</th>
<th>R-C</th>
<th>B-C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Coarse aggregate (kg/m³)</td>
<td>1164</td>
<td>1164</td>
<td>1164</td>
<td>1164</td>
<td>1164</td>
</tr>
<tr>
<td>Sand (kg/m³)</td>
<td>613</td>
<td>613</td>
<td>613</td>
<td>613</td>
<td>613</td>
</tr>
<tr>
<td>Cement (kg/m³)</td>
<td>418</td>
<td>418</td>
<td>418</td>
<td>418</td>
<td>418</td>
</tr>
<tr>
<td>Water (kg/m³)</td>
<td>205</td>
<td>205</td>
<td>205</td>
<td>205</td>
<td>205</td>
</tr>
<tr>
<td>Bacterial spores content (cells/cm³)</td>
<td>0</td>
<td>2.8 × 10⁹</td>
<td>2.8 × 10⁹</td>
<td>2.8 × 10⁹</td>
<td>2.8 × 10⁹</td>
</tr>
<tr>
<td>Calcium lactate (kg/m³)</td>
<td>0</td>
<td>12.26</td>
<td>12.26</td>
<td>12.26</td>
<td>12.26</td>
</tr>
<tr>
<td>Types of carriers</td>
<td>None</td>
<td>None</td>
<td>EP</td>
<td>RA</td>
<td>DE</td>
</tr>
</tbody>
</table>

2.4 Crack Formation

At the end of curing, concrete specimens were loaded on an electro-hydraulic servo testing machine to prefabricate cracks, as shown in Fig. 1. As the three point bending test was adopted to prefabricate cracks, the width of the crack generated gradually decreased from bottom to top. The displacement control mode was used with a loading rate of 0.05 mm/min. The formed crack widths were in the range of 0.1 to 0.4 mm. After the cracks were formed, they were removed from the press and allowed to stand for 5 to 8 hours, and the crack width was determined until the crack width was constant. That is, the initial crack width used in this study was the constant crack width when the specimen was taken out of the press and left for 5 to 8 hours. The points were marked along the crack to measure width, and the initial width was recorded. During the test, the specimens were placed on a pallet in a natural environment for watering and curing, and the entire pallet was moved to ensure that the specimens were stable. The crack width was measured at 3, 7, 14, 21, and 28 days of healing, respectively.

2.5 Self-healing effect characterization methods

To compare crack-healing capacity of specimens prepared with different carriers over time, the cracks were measured and recorded with crack width measuring instrument. The crack width measuring instrument was...
based on the principle of microscopy (see Fig. 2). The crack can be magnified 150 times by focusing and the scale value was read to accurately record the actual size. The division value is 0.005 mm, and crack observation error was less than 0.02 mm.

After 28 days of healing, to investigate the healing product formed inside the crack, the samples (1 cm × 1 cm × 2 cm) were cut along both sides of the crack with a small cutter. The specimens were winded with adhesive tape to prevent section of samples from being polluted. Next, the specimens disconnected along the crack were placed on the loading platform of a Quanta 600 FEG Scanning Electron Microscope (SEM) for analysis. Recycled aggregate at crack was also collected and subjected to Field Emission (FE-SEM) analysis to visualize the difference with the initial recycled aggregate. At the same time, analysis by Energy Dispersive Spectroscopy (EDS) was performed to determine the nature of produced precipitation. In addition, to illustrate that recycled aggregate can be used as a protective carrier to immobilize bacteria due to its high porosity, mercury intrusion porosimetry testing was performed on the new and old mortar. Recycled aggregate was selected for small diameter samples (particle size was about 6 mm) which were frozen in liquid nitrogen and subjected to cryo-vacuum evaporation for one week for pore water removal prior to mercury intrusion porosimetry (MIP) testing.

To investigate further the feasibility of recycled aggregate-immobilized bacteria as self-healing agent carrier, the surface pH and numerical tube pressure were tested. Surface alkalinity of three different carriers was measured. The alkalinity of the sample was characterized by pH. The carrier and slurry were mixed and stirred according to the mixing proportion of concrete specimens in this study, and the extract was filtered using a filter paper and tested using a pH meter (Ding et al. 2019).

Next, the strength of the different carriers was tested. The crushing index value of RA was measured in this study. Samples of 3000 g were weighed and air dried, and acicular and flaky particles that were larger than 19.0 mm and smaller than 9.50 mm were removed. The sample was divided into 2 layers and placed in a circular mold. After each layer of sample was loaded, it was turned and leveled, and the surface of the in-mold sample was covered with a pressure head. Then the circular mold with the sample was placed on a pressure testing machine and uniformly loaded to 200 kN at 1 kN/s, and stabilized for 5 s before unloading. The crushing index of recycled aggregate was obtained by the above scheme. The procedure for numerical tube pressure testing of expanded perlite and diatomaceous earth was as follows. The sample of 10 to 20 mm grain size was sieved, and the sample was loaded with a pressure cylinder. The weight of the loose material was measured three times, and the arithmetic mean was taken. The sample was weighed into the cylindrical pressure vessel according to the above sample amount, first tapped with a wooden hammer along the circumference of the tube wall, and then the guide tube and the stamping die were mounted to check the lower scale line of the stamping die and the upper side of the guiding cylinder. The pressure cylinder was placed on the lower pressure plate of the press, and was uniformly loaded at a speed of about 30 to 50 kg per second. When the stamping die was pressed to a depth of 20 mm, the pressure value was recorded.

3. Result and discussion

3.1 Healing capacity of cracks
3.1.1 Morphology of healed cracks
The cracks were magnified 150 times by crack width measuring instrument to observe more clearly the healing condition so that the width value at each crack point could be measured more accurately. Figure 3 shows the microscopic images of cracks on the surfaces of the five types of specimens at different healing times. Crack width of different concrete specimens was observed to decrease gradually over time. Healing precipitation was observed at cracks. The cracks width decreased slightly for the specimens that were directly introduced bacteria without protective carrier at 14 days of healing. However, cracks width basically no longer changed at 28 days, as shown in Figs. 3(a) and 3(b). The reason behind this phenomenon was that bacteria were hard to survive in high alkaline environment in the concrete matrix.

After 28 days of healing, marked cracks of the E-C and R-C specimens were almost completely healed. Values of completely healed crack width after 28 days of healing were up to 0.32 mm and 0.28 mm, respectively. The cracks of the B-C specimen were only partly healed (the value was up to 0.14 mm). Cracks healing capacity of R-A and E-C specimens was superior to B-C specimens. The cause of this phenomenon was that surface pore diameter of the recycled aggregate and expanded perlite was much larger than the diatomaceous earth. The pore diameter of the three protective carriers was 10 to 100 µm, 1 to 50 µm and 0.05 to 0.8 µm, respectively, leading to a large amount of adsorbed bacterial suspension in the
micropores and the increased bacterial concentration promoted the formation of precipitation. Compared to other bacteria-based concrete specimens, the cracks of N-C specimen were barely healed (the healed cracks width was 0.05 mm). The crack images are shown in Figs. 3(i) and 3(j), and it can be seen that there is no obvious healing production. The reason for the formation of a small amount of healing products was that concrete can be naturally healed by itself due to further hydration of unhydrated cement particles and precipitation of calcium carbonate crystals at the smaller width of cracks (Kastis et al. 2006).

3.1.2 Microstructure analysis of precipitate
Except for the N-C specimens, the morphology of the products observed in the cracks of all the other specimens was consistent, and the precipitate was calcium carbonate by EDS analysis, which was consistent with the theory of microbial induced carbonate precipitation (Qian et al. 2015). Healing products formed at crack surfaces of all specimens after 28 days of healing were subjected to SEM analysis to inspect their microstructure, as shown in Fig. 4. Products were found in all the cracks of the four types of specimens. Figure 4(a) shows crack-healing products of the specimens that were directly introduced bacteria without protective carrier. Few calcium carbonate crystals were observed in the cracks and were unevenly distributed. The reason could be that the survival ratio of bacteria in highly alkaline concrete decreased significantly, and internal micro-cracks caused by the load during the pre-fabrication of cracks provided small amount of space for microbial survival. As seen from Fig. 4(b), a large number of cavities were observed on the crack surface of B-C specimens, which indicated that the healing of the crack was incomplete.

Figures 4(c) and 4(d) show that the healing products were very dense and could uniformly cover the surface of the R-C and E-C specimens, which revealed that recycled aggregate and expanded perlite could provide effective protection for bacteria to promote compound formation. The difference between Figs. 4(c) and 4(d) could be observed that the diameter of crystals formed at cracks of E-C specimen was small and uniform, and the crystals were uniformly distributed in a single layer. In contrast, the crystals diameter of R-C specimens was larger, which was conducive to repair crack completely. A large amount of obvious CaCO$_3$ crystals formed only at cracks of the bacteria-based specimens, which indicated that its formation was associated with bacterial activity. Little precipitation was observed at the crack of the N-C sample by crack width measuring instrument, which was different from the case of the bacterial-based concrete specimen.

3.1.3 Crack-healing quantification
From a qualitative point of view, the healing capacity of bacteria-based concrete specimens exhibited better effect than the specimens were directly introduced bacteria without protective carrier. To compare crack-healing capacity of specimens prepared with different carriers, the actual quantification of crack-healing via crack width measuring instrument was performed in this study. In order to simulate the actual project more closely, the
prefabricated cracks width was controlled at about 0.3 mm and recorded. As the three-point bending test was adopted to prefabricate cracks, the width of the crack generated gradually decreased from bottom to top. Therefore, in this study, cracks were marked every 0.5 cm along the length to measure accurately the wide variation of the different locations for each crack. The healing ratio was calculated for each location according to Eq. (2).

\[
\delta = \frac{W_0 - W_t}{W_0} \times 100\% \tag{2}
\]

where \(w_0\) is the initial crack width and \(w_t\) is the measured width at time \(t\).

Figure 5 shows the variation of the maximum crack width for the five types of specimens at different times. Compared to D-C specimens, the healing capacity of E-C, R-C and B-C specimens was significantly enhanced, which indicated that crack repair was associated with bacterial activity. Among them, healed width in E-C and R-C specimens (0.32 mm and 0.28 mm, respectively) were the most obvious after 28 days of incubation, while the width healed in B-C specimens was only up to 0.14 mm. The crack in E-C specimens was completely healed at 28 days. At the healing time of 14 days, the D-C specimens exhibited a few crack healings, with no further healing observed over time. Healing capacity of the bacteria-based specimens was much better than other specimens due to bacterial spores were well protected from direct exposure to high alkaline environment in the concrete matrix.

In order to evaluate the healing capacity of cracks more synthetically, the average healing ratio was calculated and analyzed as shown in Fig. 6.

The average healing ratio was calculated for each type of specimens according to Eq. (3).

\[
\bar{\delta} = \frac{\sum \delta_i}{n} \tag{3}
\]

The crack-healing ratio of concrete specimens that immobilized in the protective carriers was higher than that of D-C specimens. The average healing ratio for five types of specimens was increased rapidly in earlier time of incubation. This phenomenon was also observed by Wang et al. (2016) and Jonker et al. (2010). The reason for this phenomenon was that bacterial suspension concentration was high and the calcium lactate was sufficient in the earlier healing time. In addition, bacterial spores were favorably exposed to moisture and air due to the larger initial crack width value, which promoted mineral precipitation. The average healing ratio in E-C, R-C and B-C specimens was 90.3%, 85% and 65.5% for 28 days, respectively, while the healing ratio of specimens that were directly introduced bacteria was 32%. The healing ratio of N-C specimen was only 17.2%. In fact, after 28 days incubation, most of the cracks in the E-C and R-C specimens had healed, and the healing ratio of few cracks was low [see Figs. 7(a) and 7(b)].

At the healing time of 14 days, all the crack healing ratios were concentrated between 20% and 80% [see Fig. 7(a)]. With prolongation of time, the crack healing ratios of E-C and R-C specimens were higher than 80%, while those of B-C specimens also were increased significantly. In contrast, the improvement of healing ratio for D-C specimens was not obvious [see Fig. 7(b)]. Moreover, concrete specimens using the recycled aggregate as the bacterial carrier also exhibited higher healing capacity in this study, that is, the recycled aggregate could be used as a protective carrier to achieve crack self-healing.

3.2 Characterization of the healing products

SEM images of crack-filling precipitations in R-C specimen after 28 days healing are shown in Fig. 8(b). The generated calcium carbonate shape was more clearly shown in the enlarged partial image in Fig. 8(b). Large amounts of crystals were formed at the mouth of crack. Recycled aggregate could be used as a self-healing agent carrier due to porosity of old mortar adhered to its surface (Liu et al. 2019b), as illustrated in Fig. 8(a). The calcium carbonate crystal generated in R-C specimens not only decreased porosity by filling the pore structure, but also improved the strength by bonding the particles together
To further illustrate the healing effect and determine the nature of produced precipitation, the EDS analysis was carried out on the obtained samples in the study. The EDS spectrum of the healing precipitation obtained at the cracks of R-C specimens was shown in Fig. 9. The results revealed that the massively formed precipitate at crack of R-C specimens was essentially an association of calcium, oxygen and carbon atoms, and the weight ratio among the elements closely matched with that of calcium carbonate, which suggested that mineral precipitates were calcium carbonate, as shown in Table 3. Actually, the components of precipitate formed at the cracks in other bacteria-based specimens were identical to the R-C specimens.

Table 3 The nature of precipitation.

<table>
<thead>
<tr>
<th>Specimens</th>
<th>R-C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Elements</td>
<td>Weight Percentage</td>
</tr>
<tr>
<td>C K</td>
<td>12.54</td>
</tr>
<tr>
<td>O K</td>
<td>59.04</td>
</tr>
<tr>
<td>Ca K</td>
<td>28.42</td>
</tr>
<tr>
<td>Total</td>
<td>100.00</td>
</tr>
</tbody>
</table>

3.3 Good carrier mechanism of recycled aggregate

3.3.1 Pore structure characteristics

Pore structure is a key factor in the selection of carriers. The recycled aggregate was derived from the crushed and screened waste concrete, and its composition was different from ordinary natural aggregate. Recycled aggregate was composed of old natural aggregates and old mortar attached to the surface. Old mortar was loose and porous, that is, recycled aggregate porous feature was beneficial to provide space for microbial survival (Kou et al. 2011).

To analyze the pore structure of recycled aggregate further, mercury intrusion porosimetry testing was performed on the old mortar, and the result obtained was compared with that of the new mortar. The new mortar was taken from the mortar specimen, and its composition...
Recycled aggregate was derived from waste concrete, and its quantitative information had been missing for a long time. Here, we paid more attention to pore structure, and other factors were not explored.

The parameters of the pore structures for the new and old mortar are summarized in Table 5. The total intrusion of the old mortar was larger than that of new mortar (0.1507 ml/g and 0.0834 ml/g, respectively), and the porosities of new mortar and old mortar was 17.68% and 28.63%, respectively. The data obtained showed that the pore diameter of old mortar was larger than that of new mortar. The high porosity of old mortar was related to the formation of precipitates. The pore diameter ranges of new and old mortars obtained through MIP analysis were 0.1 to 1 μm and 0.1 to 1 mm, respectively (see Fig. 10). Furthermore, the range of pore size distribution in old mortar was wide, and the ratio of 100 μm to 1 mm was the highest. The difference in the pore size of the old mortar with a wide range of fluctuation values indicated that old mortar had loose texture and uneven pores. As seen from Fig. 10, the old mortar has the maximum value of cumulative intrusion at three different pore diameters. In contrast, only one maximum was observed for the new mortar. This indicated that the new mortar has a pore size ranging from 10 nm to 100 nm and its maximum pore size was much lower than that of the old mortar.

Table 4 Mix proportion of mortar specimens.

<table>
<thead>
<tr>
<th>Component</th>
<th>(kg/m³)</th>
<th>(kg/m³)</th>
<th>(kg/m³)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mortar specimens</td>
<td>418</td>
<td>205</td>
<td>613</td>
</tr>
</tbody>
</table>

Table 5 Parameters of the pore structures.

<table>
<thead>
<tr>
<th>Component</th>
<th>Total Intrusion (ml/g)</th>
<th>Average Pore Diameter (nm)</th>
<th>Porosity (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Old mortar</td>
<td>0.1507</td>
<td>78.2</td>
<td>28.63</td>
</tr>
<tr>
<td>New mortar</td>
<td>0.0834</td>
<td>29.5</td>
<td>17.68</td>
</tr>
</tbody>
</table>

The water to cement ratio was 0.48. The mix proportion of mortar specimens is shown in Table 4. Recycled aggregate was derived from waste concrete, and its quantitative information had been missing for a long time. Here, we paid more attention to pore structure, and other factors were not explored.

The pore diameter ranges of new and old mortars obtained through MIP analysis were 0.1 to 1 μm and 0.1 to 1 mm, respectively (see Fig. 10). Furthermore, the range of pore size distribution in old mortar was wide, and the ratio of 100 μm to 1 mm was the highest. The difference in the pore size of the old mortar with a wide range of fluctuation values indicated that old mortar had loose texture and uneven pores. As seen from Fig. 10, the old mortar has the maximum value of cumulative intrusion at three different pore diameters. In contrast, only one maximum was observed for the new mortar. This indicated that the new mortar has a pore size ranging from 10 nm to 100 nm and its maximum pore size was much lower than that of the old mortar.

Additionally, DE is a soft natural siliceous sedimentation with a primary pore size between 0.05 μm and 0.8 μm, which is much smaller than that of the recycled aggregate. This was also the reason why the B-C specimen repair effect was much lower than the R-C specimen.

3.3.2 Alkalinity analysis

The activity of bacteria was greatly affected by the alkaline environment of the carrier surface. Bacterial activity decreased in high alkaline environment in the concrete matrix, resulting in low healing ratio. With the incorporation of a porous carrier, micro porous provided microorganism more space for precipitation. Therefore, it is significant to study the pore structure and alkaline environment of the carrier for self-healing concrete. Bacillus pasteurii used in this study could survive in the environment of pH 6 to 11, and could show optimal activity in the environment of pH 9; that is, survival of bacteria beyond pH 11 was very difficult. When pH = 13, the bacteria were still active, but there was no denying that their survival ratio decreased greatly. In other words, bacterial growth was inhibited by the high alkalinity. In terms of crack healing, R-C was superior to N-C. The reason behind this phenomenon was that a large amount of old mortar attached to the surface of RA was carbonized for a long time, causing to the surface alkalinity of RA to decrease. In the process of the concrete carbonization, the content of Ca(OH)₂ in the capillary pores of concrete gradually decreased, which inevitably reduced the alkalinity of interior and surface. The carbonation process of calcium hydroxide occurred in old mortar according to the following reaction:

\[
\text{Ca(OH)}_2 + \text{CO}_2 = \text{CaCO}_3 + \text{H}_2\text{O}
\]

The pH value of ordinary concrete is 12 to 13, and the value after carbonization obviously decreased (Sun et al. 2011). On the other hand, massive micropores on the surface of the RA were fully exposed to air due to the formation of cracks. The diffusion path of CO₂ increased in concrete, leading to a faster rate of carbonization, which is also an important reason for the low alkalinity of the recycled aggregate. The pH value of the concrete was the same as that of the concrete specimen used in this study. The water to cement ratio was 0.48. The mix proportion of mortar specimens is shown in Table 4. Recycled aggregate was derived from waste concrete, and its quantitative information had been missing for a long time. Here, we paid more attention to pore structure, so other factors were not explored.

The parameters of the pore structures for the new and old mortar are summarized in Table 5. The total intrusion of the old mortar was larger than that of new mortar (0.1507 ml/g and 0.0834 ml/g, respectively), and the porosities of new mortar and old mortar was 17.68% and 28.63%, respectively. The data obtained showed that the pore diameter of old mortar was larger than that of new mortar. The high porosity of old mortar was related to the adsorption ratio of the bacterial suspension, that is, with the increase of porosity, the adsorption for bacterial suspension was increased, which further promoted the formation of precipitates.

The pore diameter ranges of new and old mortars obtained through MIP analysis were 0.1 to 1 μm and 0.1 to 1 mm, respectively (see Fig. 10). Furthermore, the range of pore size distribution in old mortar was wide, and the ratio of 100 μm to 1 mm was the highest. The difference in the pore size of the old mortar with a wide range of fluctuation values indicated that old mortar had loose texture and uneven pores. As seen from Fig. 10, the old mortar has the maximum value of cumulative intrusion at three different pore diameters. In contrast, only one maximum was observed for the new mortar. This indicated that the new mortar has a pore size ranging from 10 nm to 100 nm and its maximum pore size was much lower than that of the old mortar.

Fig. 10 Pore size distributions of old mortar (a) and new mortar (b).
with RA, DE and EP was measured to be 9, 8.5 and 7.5, which indicates that the concrete with RA is more suitable for the Bacillus pasteurii used in this study. The survival ratio of bacteria was increased in this alkaline environment, and the mineralization efficiency was improved.

3.4 Application advantages and prospects
Recycled aggregate was first used as a novel self-healing carrier and exhibited good healing effect. The maximum crack width healed was near 0.28 mm, and the healing ratio of most cracks almost reached up to 100% after 28 days of healing. Except for the porosity and alkalinity, good mechanical properties (Xiao et al. 2004; Liu et al. 2019a, 2019b, 2019c) should be considered during the choice of carriers, so that the strength of the concrete was not affected by the carriers.

The crushing index of recycled aggregates and the numerical tube pressure of lightweight aggregates such as expanded perlite and diatomaceous earth were tested. The crushing index of recycled aggregate was 17%. Numerical tube pressure of the expanded perlite and diatomaceous earth used in this study was 0.18 MPa and 0.49 MPa, respectively. It is obvious that the mechanical properties of recycled aggregates are better than lightweight aggregates. Recycled aggregate is more conducive to improve strength of the self-repairing concrete.

From the perspective of the economy, RA is derived from the waste concrete of construction waste, which means that subsidies ($5 to $10 per m3) are given by the government when recycled aggregates are used. For recycled aggregates, there is a huge market in China, where the annual production of recycled aggregates accounts to about 45% of the world's total. On the contrary, the cost of EC and DC particles reaches up to $15 to $20 per m3 and $200 to $280 per m3, respectively, in Xi'an, Shaanxi province, and will change as policy changes in the future. The feasibility of using recycled aggregate in self-repairing concrete is improved due to its superior economic benefits. At the same time, the increase in the utilization rate of recycled aggregate has contributed to environmental protection.

4. Conclusions
In this study, Bacillus pasteurii was used as self-healing agent and the RA was used as a protective carrier to prepare concrete specimens. The feasibility of RA immobilizing bacteria for the cracks self-healing was investigated. The healing capacity of this technique was compared with three other incorporation techniques, i.e., direct introduction of bacteria, DE-immobilized bacteria and EP-immobilized bacteria. The main conclusions drawn are outlined below.

The healing capacity of R-C specimens is superior to specimens directly introduced bacteria without protective carrier. The value of completely healed crack width after 28 days of incubation was up to 0.28 mm, which is larger than the value of 0.07 mm for specimens that were directly introduced bacteria. The healed crack width value of R-C specimens was close to that of E-C specimens (0.28 mm and 0.32 mm, respectively), and larger than that of B-C specimen (the value reached up to 0.14 mm).

EDS analysis confirmed that the weight ratio of Ca/C/O is 1:1:3, and the precipitation formed at the cracks was all calcium carbonate, which was consistent with the mechanism of microbial induced carbonate precipitation.

Recycle aggregate was investigated to effectively protect bacteria in harsh conditions of concrete. This is attributed to the porosity of the surface old mortar and the low alkalinity formed by long-term carbonization, leading to high adsorption and survival rate of the bacterial cells.

The mechanical properties of RA are higher than that of the other two carriers, which provides a possibility for improving strength of self-healing concrete. The characteristics of low cost and environmental protection make RA more promising than EC and DE particles in the application for protective carriers.

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References
610-619.
recycled concrete aggregate - a review.” Journal of Cleaner Production, 221(1), 466-472.