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Microbial technique to treat recycled aggregates from construction waste for its effective reutilization in concrete

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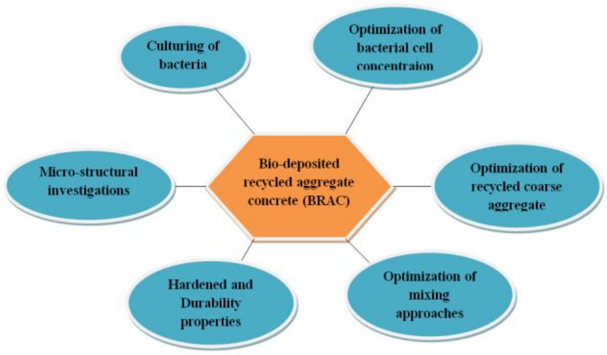
Graphical abstract



Abstract

Excessive consumption of natural resources for concrete production results in the diminution of conventional resources, leading to the scarcity of construction materials. Perhaps, the dumping of construction wastes increases disposal problems. Nevertheless, the requirement for aggregates in the construction sector increases. The recycling of construction waste to produce recycled coarse aggregate (RCA) as a suitable alternative to natural coarse aggregates (NCA) conserves natural resources and promotes sustainability in construction. However, the quality of recycled coarse aggregate was inferior compared to natural coarse aggregates due to the adherence of mortar. This paper investigates the sustainable use of Bacillus subtilis with different concentrations to enhance the quality of RCA. The concrete mixes manufactured with optimized bio-deposited recycled coarse aggregate (BRCA) were tested for their mechanical and durability properties. It could be observed that the strength of bio-deposited recycled aggregate concrete (BRAC) was enhanced by 12.63% relative to normal aggregate concrete (NAC), and the durability properties such as water absorption, chloride penetration and carbonation of BRAC were reduced by 18.53%, 16.52% and 20% relative to recycled aggregate concrete (RAC). Microstructural studies through SEM and XRD revealed the deposition of $CaCO_3$ on the micro-pores of RCA, and that improves the properties of the concrete.

Keywords: Bacillus subtilis, recycled aggregate concrete, bio-deposition, $CaCO_3$, sand enveloped mixing approach.

1. Introduction

Construction & Demolition (C&D) wastes resulting from the retrofitting of concrete structures were generated in huge quantities annually and dumped in landfills. Subsequently, almost 800 billion tons of natural resources are utilized by industrialists as ingredient to manufacture concrete in 2017 and which they anticipated would rise the generation of demolition waste to 270 billion tonnes in next 10 years (Schandl *et al.,* 2017). The concrete production rose to 4.4 billion tonnes in 2022 and anticipated to increase over 5.5 billion tons in 2050. Almost one-third of such manufacturing ends up as C&D waste. The central pollution control board (CPCB) of India estimated that around 65 million tons of municipal wastes are generated yearly, of 62% comprises of recyclables such as wood, concrete, glass etc. Among them, only 75% were collected and out of which 20 to 30% were being processed (TERI, 2022). European Aggregate Association (EAA) estimated the requirement of 4000 tonnes of aggregates for constructing a residential villa and 3000 tonnes for laying one kilometre of a road. From the statistical data, it is observed that the use of conventional resources as construction material and the generation of C&D wastes were increasing simultaneously. The former will cause an imbalance in the ecology by diminution of natural resources, while the latter will affect the integrity of the environment. Thus C&D waste has to be used as an appropriate alternative for NCA in the concrete. However, the smearance of cement mortar on RCA affects the concrete properties, hence restraining its use in the concrete. Suitable treatments for RCA will tend to enhance the quality of RCA by eradicating cement mortar or coating it with an impermeable medium.

Several research have been done with the RCA as an effective replacement to NCA in concrete. Puthussery *et*

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al. (2017) reported that beyond 25% substitution of RCA, the strength of RAC was reduced ensuing from the poor quality of the RCA, adhered mortar, and void packing nature of the RCA. Andal *et al.* (2016) utilized both preserved and unpreserved RCA in the study and observed replacement beyond 30% shown negative influence on the concrete properties, however the preserved RCA performed better compared to unpreserved RCA. Lavado *et al.* (2020) found that for the RAC mix with lower w/c ratio (0.35 to 0.4), addition of superplasticizers shown higher slump loss whereas at 0.5 w/c ratio, slump loss was only 6% at first 30 minutes and gradually increased to 63% at 90 minutes. Arunkumar *et al.* (2023) observed that optimizing polypropylene aggregate to 15% with recycled water increased the strength equivalent to the control concrete and replacement beyond 15% reduced its strength. Naveen Arasu *et al.* (2023) used recycled nano-material and observed that the strength was compressive enhanced by 32% and tensile strength was enhanced by 24% and 0.04% replacement. *Mavroulidou et al.* (2010) investigated the possible utilization of rubber tyre as aggregates in concrete and observed that the strength was nearly 95% of control concrete when used as coarse aggregate and 89% when used as fine aggregate at 28 days. The lower specific gravity of rubber tyre reduces the strength of the concrete.

Mi et al. (2020) reported that RCA collected from the source concrete with higher strength shown improved strength and carbonation resistance with ratio ranging from 0.6 to 1.1 for the former and 11 to 8 for the latter. *Mwasha et al.* (2018) optimized the particle packing of pre-soaked RCA and observed a highest compressive strength of 80 MPa with the influence of mineral admixtures. The combined effect of pre-soaking, particle packing optimization and mineral admixtures shown better influence on the concrete properties. It could be observed that the addition of RCA tend to disturb the concrete properties, limiting its utilization. Several treatments were developed to RCA with acids, polymers, slurries, carbonation, microbes etc. to enhance its quality.

Earlier studies with bacteria tend to improve the concrete properties through bio-deposition of $CaCO_3$ (Bachmeier *et al.* 2002; Park *et al.* 2010; Majumdar *et al.* 2012). (Krishnapriya *et al.* 2016) isolated *Bacillus megatherium*, *Bacillus licheniformis,* and *Bacillus flexus* and observed higher strength with all three species than control concrete due to the $CaCO_3$ precipitation and among the three, *Bacillus flexus* performed better due to its 100% higher similarity index with the parental species. Mondal *et al.* (2017) used *Bacillus Cereus* and *Bacillus Subtilis* at various concentrations and observed that the concrete with 10^3cells/ml of *Bacillus Cereus* and 10^5cells/ml *Bacillus Subtilis* of 10^5 cells/ml shows higher strength owing to the higher calcite precipitation by microbes that seals up the micro-pores in the concrete. *Nain et al.* (2019) used 0.8 x 10^9cells/ml concentration of *B. Subtilis* and *B. Megaterium* and found that strength of the RAC was improved around 15% with both bacterial species. The microstructure revealed dense rhombohedra-shaped calcite crystals that deposit on the cracked surface and heal it. Thus it could be observed that microbes has the ability to improve concrete properties by sealing pores/voids in concrete through $CaCO_3$ deposition. The same technique was implemented to augment the quality of RCA to check its suitability in the concrete.

Grabiec et al. (2012) used Sporosarcina pasteurii to enhance the RCA and found that the perviousness of RCA was decreased, and it is more efficient with smaller fractions of RCA. *Wu et al.* (2020) inferred that the rate of absorption of water by RAC was reduced by 28.3%, and the strength of RAC was enhanced by 16.1% with the bio-deposition of RCA. The microbes precipitate the CaCO3 crystals that seal the cracks on the cement mortar adhered to RCA. However, *Zhan et al. (2019*) adopted microbial treatment to finer fractions of RCA and found a 32% improvement in the strength even with 100% of RCA. The solidification of the micro-structure through $CaCO_3$ precipitation augments the properties of the RAC. *Zhu et al.* (2019) observed that microbe-induced CaCO3 precipitation promotes the formation of CSH, CH, Aft, and AFm, which fill the cracks on the RCA and increase its strength. *Wu et al.* (2018) developed an alternative technique with microbes through respiration and observed improved RCA properties, thereby enhancing RAC properties. However, fewer studies were performed to examine the influence of bio-treated RCA in the concrete mixture to study its mechanical and durability properties. This study used *Bacillus subtilis* as a microbe to enhance the quality of RCA and properties of RAC.

2. Experimental work

2.1. Materials

This study utilizes 43 grades ordinary portland cement, river sand as natural fine aggregate (NFA) and river gravel as natural coarse aggregate. Construction wastes were procured from Structural Engineering Research Center (SERC), India, and only the concrete segments were sampled and used as RCA. The collected samples were crushed and sieved to fractions ranging from 4.75-mm to 20 mm to eliminate finer dust particles. The properties of aggregates determined following IS 2386 (Part III)-1963 were given in Table 1. Table 1 shows the inferior properties of the RCA owing to its higher absorption, crushing index and less density than NCA as a result of smearance of mortar on RCA (*Sivamani et al. 2020, Sivamani et al. 2021a; Abrahams et al. 2018*). The photographic and microscopic observations of collected NCA and RCA samples are shown in Figures 1 & 2. It could be observed that the RCA particles are highly angular, with cementitious particles smeared on their surface. This is due to the repeated crushing of RCA to reduce the particles to size from 4.75-mm to 20 mm. The microscopic observation reveals the cement mortar remains on RCA and was visible through the loose irregular particles, whereas the images of NCA show fewer angular dense particles. The loose irregular RCA particles possess micro-cracks that increase the porosity of RCA. The XRD images

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of NCA and RCA are shown in Figure 3. The peak in NCA pattern specify the incidence of SiO_2, $NaAlSiO_3O_8$, and $CaCO_3$, with the highest being SiO_2. This could be favourable in the development of a C-S-H that promotes strength in the concrete. In RCA, the peak signify the calcite compound ensuing from the cement mortar and traces of SiO_2.

Figure 1. Visual observation (a) NCA (b) RCA

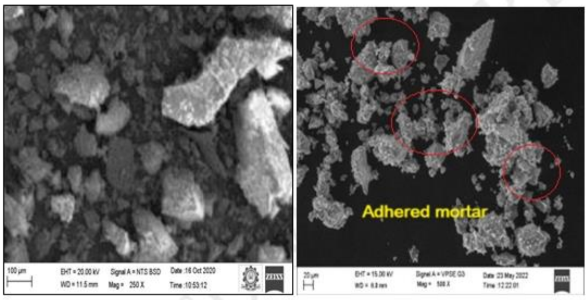

Figure 2. Microscopic observation (a) NCA (b) RCA

2.2. Bio-deposition treatment

The genetic variant of the *Bacillus subtilis* was obtained from a Gene bank in Mumbai, India (Gene ID: Chandigarh/ NCBI/EMBL/DDBJ/2341). The technique to culture *Bacillus subtilis* was performed with reference to the procedure (Sahoo *et al.* 2016; González *et al.* 2017). The strains of the *Bacillus subtilis* were preserved in a medium comprising 30 g/l urea and an extract of yeast (Y.E) each. The Y.E was diluted in 1000 ml of water, pasteurized at 110^o C for 30 minutes, and cooled at room temperature. The urea was mixed in 0.1 liters of distilled water and diverse with pasteurized Y.E. The collected bacterial strains were put into the diluted solution and centrifuged at 200 rpm for 24 hours. It is then diluted to attain 10^1, 10^3, 10^5, and 10^7cells/ml concentration bacteria. The diluted concentrations of the bacterial strain were quantified by the optical density test method. Figure 4 depicts the cultivation of microbes, and the cultivated microbial solution.

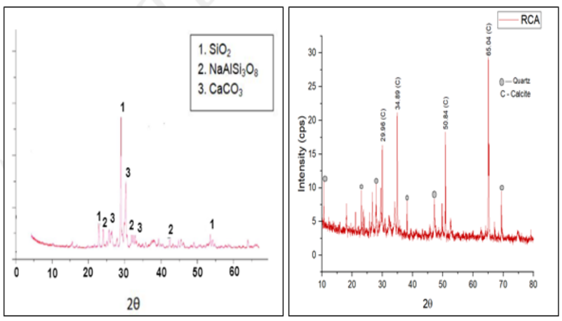

Figure 3. XRD patterns (a) NCA (b) RCA

The biotreatment to RCA was performed at room temperature with R.H >60%. The RCA was soaked in the bacterial culture of 10^1,10^3,10^5 cells/ml and 10^7cells/ml concentration for 24 hours. The aggregates are then immersed in a bio-deposition medium containing 0.5 M of urea and calcium nitrate each for three days. After three days, the samples of the RCA were taken out, cleaned, and desiccated and used as bio-deposited RCA (BRCA) in the concrete.

Figure 4. Cultivation of microbes

2.3. Concrete mixtures

The concrete mix was prepared in 1:1.92:3.11 with 0.45w/c ratio as per IS 10262 (2009). The concrete mix was labelled as BR/R-x-N/S, where 'BR' represents the biotreated RCA, 'R' represents the RCA, 'x' represents the percentage of RCA and 'N/T' represents the normal mixing approach (NMA) / sand-enveloped mixing approach (SEMA). Table 2 depicts the quantity of concrete ingredients used in the study. The NCA and RCA were saturated in prior to achieve surface saturated dry (SSD) to avoid excess water absorption during the mixing of the concrete. The cementitious particles smeared on the RCA soak up additional water while mixing, affecting the workability of the concrete. Henceforth, the concrete mixes were manufactured with SEMA technique that improves the workability of the RAC (Liang *et al.* 2015). Figure 5 depicts the illustration of NMA and SEMA.

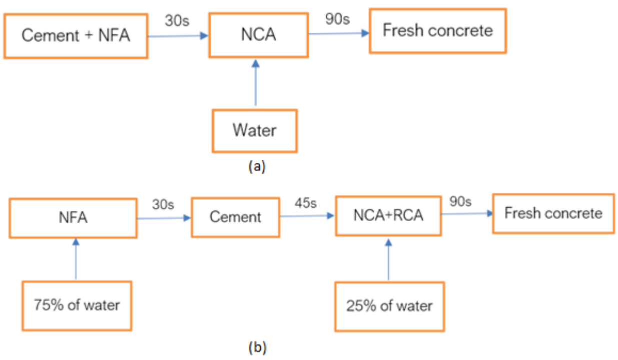

Figure 5. Schematic Illustration (a) NMA (b) SEMA

2.4. Testing methodology

The bio-treated aggregates used in the study were tested for its suitability by performing water absorption, crushing index and density as per IS 2386 (Part III)-1963. A whole of

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3 mix series were prepared with NCA, RCA and BRCA. The concrete mixes are manufactured by NMA and SEMA (Figures 3 and 4) and casted into cubes (150 mm), cylinders (150 mm x 300 mm) and prisms (500 mm x 100 mm x 100 mm). The fresh mixes were tested for its workability as per IS 1199 (1959) and the hardened concrete were tested for strength and durability properties. The concrete cubes and cylinders were tested for its compressive (CS) and tensile strength (TS) as specified in ASTM C39/ C39-M. The concrete prisms were tested for its flexural strength (FS) as specified in ASTM C469. All the hardened property tests were performed in trios at 7 and 28 days.

Table 1. Aggregate properties

Aggregate	Specific Gravity	Water absorption (%)	Relative density (kg/m^3)	Crushing index (%)
NFA	2.61	0.12	1651	19.43
NCA	2.74	0.87	1976	20.41
RCA	2.41	6.13	2431	25.23

Table 2. Concrete mix proportions

Mix ID	(Kg/m^3)					
	Cement	NFA	NCA	RCA	BRCA	Water
R-0-N	400	799	1029	0	0	180
R-0-S	400	799	1029	0	0	180
R-30-N	400	799	720	309	0	183
R-30-S	400	799	720	309	0	183
R-100-N	400	799	0	1029	0	190
R-100-S	400	799	0	1029	0	190
BR-30-S	400	799	720	0	309	180
BR-100-S	400	799	0	0	1029	180

The concrete cubes were tested for its water absorption (WA) at 28 days as specified in ASTM C1585. Initially, dry samples were weighed and soaked in water for 28 days under laboratory temperature. The samples were then taken out, allowed to dry and balanced and the variance in balance before and after soaking depicts the water absorption. The concrete cubes were used to measure the sorptivity as specified in BS 1881 (208). The carbonation resistance (CR) were determined at 7 and 28 days (*Geng et al. 2013*). The cubes cured at room temperature were accelerated with 25% CO_2 concentration. The carbonated samples were then cut and sprayed with phenolphthalein indicator. The chloride penetration (CP) of the samples were tested at 28 and 90 days as per ASTM C1202 (2019).

3. Results and discussion

3.1. Material properties

Table 3 depicts the properties of aggregates used in the concrete mixes. The significant change in water absorption of BRCA could be observed, whereas the change in relative density and crushing index was not significant. The relative density of BRCA was only 2.87% lower than the of RCA, and the crushing index of BRCA was only 8.6% lower than RCA. The effect of $CaCO_3$ precipitation was much reflected in water absorption compared to the relative density and crushing index (Liu *et al.* 2020). This is due to the insignificant change in the mass and volume of BRCA after $CaCO_3$ precipitation. The water absorption of BRCA is 33% lower than RCA. This is due to the $CaCO_3$ precipitation that fills the micro-pores and hence reduces the water absorption of RCA (Grabiec *et al.* 2012, Zhu *et al.* 2019, González *et al.* 2017, Qiu *et al.* 2014).

3.2. Characterization studies on aggregates

Figures 6 and 7 show the SEM and XRD images of RCA and BRCA used in the study. From the SEM images, it is observed that the surface of BRCA was deposited with $CaCO_3$ precipitates, and the surface of the RCA was deposited with loose cement particles. Improved magnification of 5µm clearly shows the $CaCO_3$ precipitation on the RCA. The bio-deposits tend to seal the micro-cracks on the porous RCA, hence improving the quality of RCA. From XRD images, it is observed that the intensity of calcite precipitation for BRCA is 80000 cps and for URCA is 35000 cps. The maximum rate of $CaCO_3$ precipitation was evident in BRCA due to the microbial activity. The Ca^{2+} ions in the medium deliver appropriate positive ion source for urea hydrolysis, promoting $CaCO_3$ precipitation. Despite this, the calcite on the RCA was evident owing to the loose mortar on its surface.

3.3. Effect of bio-deposition on the properties of RCA

The influence of bio-deposition on the properties of RCA was evaluated through weight increase, and water absorption is shown in Table 4. The increase in cell concentration increases the weight of the sample and decreases its water absorption. This is because as the bacterial concentration increases, the rate of $CaCO_3$ precipitation increases. The precipitated $CaCO_3$ crystals seal the pores on RCA, increasing its mass and decreasing the porosity. The optimal concentration of 10^5cells/ml was observed as beyond which it increases the water absorption. At optimal concentration, $CaCO_3$ precipitate percolates inside the matrix and seals the surface whereas at 10^7cells/ml $CaCO_3$ precipitate seals the surface rather than percolation into the matrix (*Jagan et al. 2022*).

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Table 3. Properties of aggregates

S. No	Aggregates	Water absorption (%)	Crushing index (%)	Relative density (kg/m^3)
1	RCA	6.3	26	2490
2	BRCA	4.2	23	2470
3	NCA	0.9	21	2200

Table 4. Variation in mass and water absorption of BRCA

S. No	Concentration (cells/ml)	Mass increase (%)	Water absorption (%)
1	10^0 (RCA)	0.38	6.27
2	10^1	0.41	5.71
3	10^3	0.47	5.18
4	10^5	0.7	4.63
5	10^7	0.93	4.92

3.4. Optimization of RCA

Figure 8 depicts the compressive strength of RAC at 28 days. The ideal replacement of RCA was observed to be 30%. The strength of R-10-N and R-30-N was 3.9% more than R-0-N at 28 days. The trivial strength enhancement was due to the angularity of RCA ensuing from the recycling process (*Jagan et al. 2020; Sivamani et al. 2021a*). The strength of R-50-N was reduced by 11.42% and the strength of R-100-N was reduced by 20.06% at 28 days. The four phase system of RAC with two ITZ, the weaker zone amongst old ITZ and new ITZ owing to higher porosity of RCA affects the concrete strength (*Thomas et al. 2018; Jagan et al. 2022*).

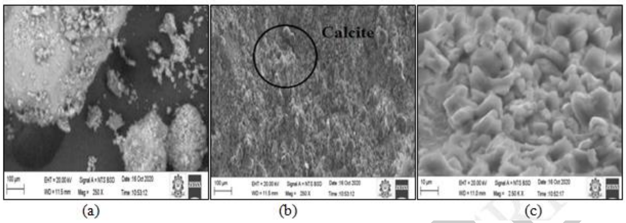

Figure 6. SEM images (a) RCA (100µm) (b) BRCA (100µm) (c) BRCA (10µm)

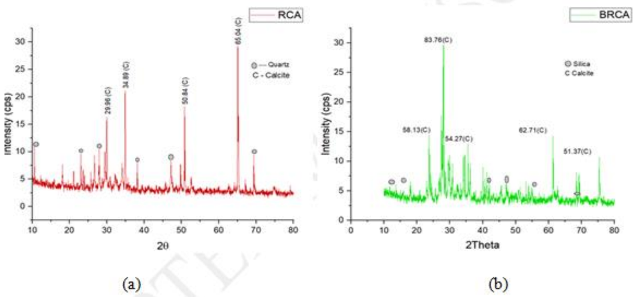

Figure 7. XRD patterns (a) RCA (b) BRCA

3.5. Mechanical properties with optimized RCA

Figure 9(a) shows the variation in CS of the optimized mixes with respect to control mix. The CS of R-0-S is 0.33% more than R-0-N at 28 days. The CS of R-30-N is 1.08% more than R-0-N, while the CS of R-30-S is 4.75% more than R-0-N at 28 days. The minor improvement in the former mixis due to the increase in angularity of the RCA owing to crushing (*Sivamani et al. 2021b; Kirthika et al. 2020*), while in the latter mix, the SEMA coats the RCA with firm mortar that reduces its water absorption during mixing (*Liang et al. 2021; Sivamani et al. 2021b; Sivamani et al. 2022*). However, the CS of R-100-N is 12.41% more than R-0-N, but the CS of R-100-S is only 9.2% lesser than R-0-N. Through bio-deposition, the CS of BR-30-S and BR-100-S was 8.17% and 12.63% more than R-0-N. Also, the CS of BR-30-S was 19.6% and 16.6% more than R-100-N and R-100-S; CS of BR-100-S was 23.5% and 20.65% more than R-100-N and R-100-S at 28 days. The $CaCO_3$ precipitation by microbes lids the RCA surface, reducing its porosity and improves the concrete strength (*Sivamani et al. 2021a; Wang et al. 2017; Wu et al. 2018*).

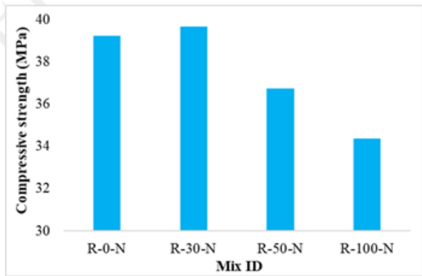

Figure 8. Compressive strength of RAC at 28 days

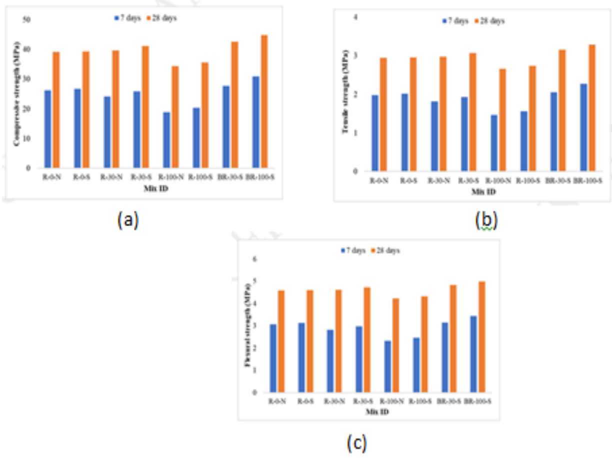

Figure 9. (a) Variation in CS (b) Variation in TS (c) Variation in FS

Figure 9(b) shows the variation in TS of the optimized mixes with respect to control mix. The TS of R-0-S is 0.26% more than R-0-N at 28 days. The TS of R-30-N is 0.87% more than R-0-N, while the TS of R-30-S is 3.83% more than R-0-N at 28 days. However, the TS of R-100-N is 9.89% lesser than R-0-N, but the TS of R-100-S is only 7.31% lesser than R-0-N. Through bio-deposition, the TS of BR-30-S and BR-100-S was 6.62% and 9.93% more compared to R-0-N. Also, the TS of BR-30-S was 15.82% and 13.29% more than R-100-N and R-100-S; TS of BR-

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100-S was 19.14% and 16.71% more than R-100-N and R-100-S at 28 days.

Figure 9(c) shows the variation in FS of the optimized mixes with respect to control mix. The FS of R-0-S is 0.20% more than R-0-N at 28 days. The FS of R-30-N is 0.66% more than R-0-N, while the FS of R-30-S is 2.93% more than R-0-N at 28 days. However, the FS of R-100-N is 7.76% lesser than R-0-N, but the FS of R-100-S is only 5.7% lesser than R-0-N. Through bio-deposition, the FS of BR-30-S and BR-100-S was 5.06% and 7.91% more compared to R-0-N. Also, the FS of BR-30-S was 12.41% and 10.53% more than R-100-N and R-100-S; FS of BR-100-S was 15.03% and 13.22% more than R-100-N and R-100-S at 28 days.

3.6. Durability properties with optimized RCA

Figure 10(a) shows the WA of the optimized mixes at 28 days. The WA of R-0-S is 2.64% lesser than R-0-N at 28 days. The WA of R-30-N and R-30-S is 14.28% and 12.76% more than R-0-N. Similarly, the WA of R-100-N and R-100-S is 21.02% and 19.83% more than R-0-N. The increase in WA of RCA owing to the adhered mortar increases the WA of the RAC (*Thomas et al. 2018; Sivamani et al. 2021a*). The WA of the hardened concrete was influenced by the pore structure of the concrete. The bio deposition treatment blocks the micro-pores on the RCA surface through the precipitation of $CaCO_3$ crystals (*Grabiec et al. 2012; Qiu et al. 2014*). The WA of BR-30-S and BR-100-S is only 9.22% and 1.82% more compared to R-0-N. Also, the WA of BR-30-S and BR-100-S is 13% and 19.58% lesser than R-100-N and the WA of BR-30-S and BR-100-S is 11.87% and 18.53% lesser than R-100-S at 28 days.

Figure 10(b) shows the sorptivity of the optimized mixes at 28 days. The sorptivity of R-0-S is 1.71% lesser than R-0-N at 28 days. The sorptivity of R-30-N and R-30-S is 34.9% and 32.91% more than R-0-N. Similarly, the sorptivity of R-100-N and R-100-S is 38.9% and 37.73% more than R-0-N. The sorptivity is the capillary uptake of water which is influenced by the pores in the hardened concrete. The higher RCA content increases the pore volume in RAC and as a result sorptivity increases (*Kirthika et al. 2020*). The bio-treatment to RCA reduces the pore volume in RAC by reducing the water absorption of RCA and thus reducing the sorptivity. The sorptivity of BR-30-S and BR-100-S is only 6.64% and 1.26% more than R-0-N. Also, the sorptivity of BR-30-S and BR-100-S is 34.52 and 38.08% lesser than R-100-N and the sorptivity of BR-30-S and BR-100-S is 33.30% and 36.93% lesser than R-100-S at 28 days.

Figure 10(c) shows the CP of the optimized mixes at 28 days. The CP of R-0-S is 1.06% lesser than R-0-N at 28 days. The CP of R-30-N and R-30-S is 7.84% and 6% more than R-0-N. Similarly, the CP of R-100-N and R-100-S is 19% and 18.26% more than R-0-N. The higher pore volume in RAC due to increase in the RCA increases the entry of Cl^- ions and affects the concrete. The CP of BR-30-S and BR-100-S is only 4.08% and 2.09% more than R-0-N. Also, the CP of BR-30-S and BR-100-S is 15.51% and 17.24% lesser than R-100-N and the CP of BR-30-S and BR-100-S is 14.78% and 16.52% lesser than R-100-S at 28 days. The bio-treatment reduces the water absorption of RCA, reducing the pore volume in RAC and thus decreasing the Cl^- penetration.

Figure 10(d) shows the carbonation of the optimized mixes at 28 days. The carbonation of R-0-S is 3.84% lesser than R-0-N at 28 days. The carbonation of R-30-N and R-30-S is 13.3% and 10.34% more than R-0-N. Similarly, the carbonation of R-100-N and R-100-S is 23.52% and 22.15% more than R-0-N. The higher pore volume in RAC due to increase in the RCA increases the entry of CO^{2-} ions and increases the rate of carbonation in RAC (*Guo et al. 2018; Silva et al. 2015*). The carbonation of BR-30-S and BR-100-S is only 7.14% and 4.41% more than R-0-N. Also, the carbonation of BR-30-S and BR-100-S is 17.64% and 20% lesser than R-100-N and the carbonation of BR-30-S and BR-100-S is 16.16% and 18.56% lesser than R-100-S at 28 days. The bio-treatment reduces the water absorption of RCA, reducing the pore volume in RAC and thus decreasing the CO^{2-} penetration.

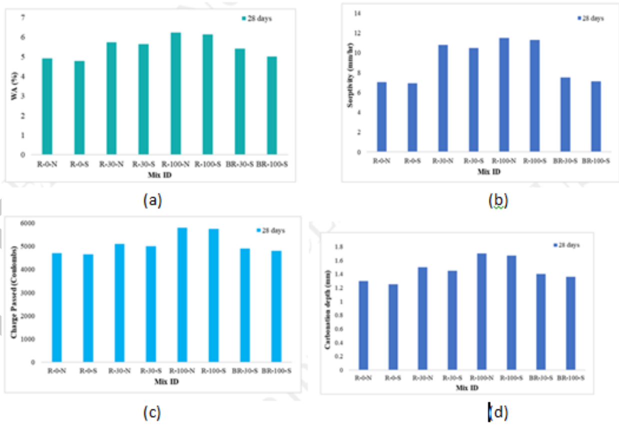

Figure 10. (a) WA of the optimized mixes (b) Sorptivity of the optimized mixes (c) RCPT of the optimized mixes (d) Carbonation of the optimized mixes

4. Discussions

The effect of bacterial treatment on the properties of URCA was assessed through weight increase, water absorption, and crushing index. It is inferred that bio-deposition increases the mass of the aggregates and reduces the water absorption and crushing index. The RCA used in the study was thoroughly washed and pre-saturated before bacterial treatment to remove the finer fractions adhered to the RCA surface (Wang *et al.* 2017). *Bacillus subtilis* was cultivated to investigate the bacterial species with optimal cell concentration for aggregate treatment. The increase in cell concentration increases bacterial precipitation in the order of 10^5-10^7cells/ml. Qui *et al.* (2014) inferred that incremental cell concentration improves the urea hydrolysis and the nucleation sites of $CaCO_3$ precipitation. This in turn increases the $CaCO_3$ precipitation and reduces the water absorption of BRCA. The influence of SEMA to enhance to quality of concrete mix tend to exhibit minor improvement in the concrete properties. In SEMA, 3/4th of the water added with NFA and cement produces stiff matrix that wraps the RCA added henceforth. The stiff wrap around RCA reduces the

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absorption of mixing water by RCA and thus improves the workability and strength of RAC (*Liang et al. 2015, Sivamani et al. 2021a*).

The improvement in the properties of BRCA eventually improves the properties of BRAC. The urea in the culture medium hydrolyse into NH_4 and CO^{2-} ions. The CO^{2-} ions reacts with $Ca(OH)_2$ to precipitate $CaCO_3$ that deposits on the RCA. The higher $CaCO_3$ precipitation on the RCA firmly accumulates the micro-cracks on the RCA that resisted the shear exerted during the concrete mixing.

In RAC, interfacial transition zone (ITZ) could be evident between RCA, fresh matrix and old matrix (*Liu et al., 2020; Wu et al., 2020*). The ITZ between fresh and old matrix becomes the weakest link due to the smeared mortar on the RCA. The $CaCO_3$ precipitated as a result of bio-deposition seals the micro-cracks on ITZ, endorsing hydration in ITZ and thus densified microstructure is formed enabling strength improvement in the RAC. Such microbial activity of *Bacillus Subtilis* improves the hardened properties of the RAC. The WA of BR-100-S was found to be lower than R-100-S and equivalent to R-0-N. When RCA was used, the pore volume of the concrete was increased that ultimately increases the WA of the RAC. Through bio-deposition, $CaCO_3$ lids the micro-cracks on RCA, eventually reducing the pore volume in RAC and hence WA of BR-100-S was equivalent to R-0-N (*Liu et al. 2020, Wang et al. 2017*). Other durability properties discussed in the study were direct reliant on the pore volume of the RCA, as which improvement was observed in bio-deposited mixes.

The effect of $CaCO_3$ precipitation on improving the properties of RAC was investigated through SEM. Figure 11 shows the SEM images of R-0-N, R-100-N and BR-100-S. It is observed that the mortar adhered to the RCA was highly porous, affecting the concrete's properties. Upon bio-deposition, dense $CaCO_3$ crystals were precipitated and deposited on the surface of the RCA (Grabiec *et al.* 2012; Wu *et al.* 2020; Liu *et al.* 2020; Wang *et al.* 2017).

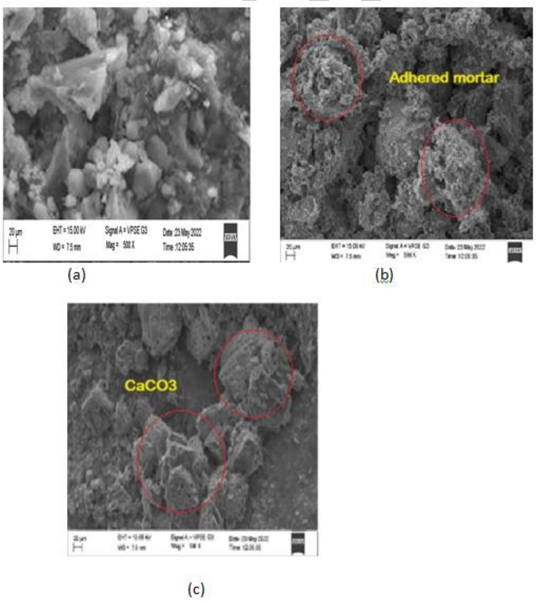

Figure 11. SEM images (a) R-0-N (b) R-100-N (c) BR-100-S

5. Conclusion

The experimental investigation on the properties of bio-deposited recycled aggregate concrete was performed. The study was performed to evaluate the properties of NCA, RCA and BRCA for its utilization in the concrete. The optimized concrete mixtures was evaluated for its hardened properties and durability properties at suitable ages. The microstructure investigations to evaluate the precipitation of $CaCO_3$ crystals by bacterial species were also conducted. The resulting inferences were made as follows:-

1. The $CaCO_3$ precipitation through urea hydrolysis by *Bacillus subtilis* reduces the RCA's water absorption.

2. Higher microbial concentration rises the $CaCO_3$ precipitation. This was evident through the increase in the mass and decrease in the absorption of BRCA.

3. The smearance of mortar on RCA decreases the replacement of RCA optimizing it to 30%, beyond which it results in strength reduction of RAC.

4. The strength of mixes with 30% of RCA, 30% of BRCA and 100% of BRCA was 4.75%, 8.17% and 12.63% more than control concrete while the strength of mixes with 100% of RCA was 9.17% lesser than R-0-N at 28 days.

5. The optimized concrete mixes with 100% of BRCA through shows improved durability due to the $CaCO_3$ precipitation and the densification of ITZ through SEMA.

The research provides an ideal solution to improve the RCA quality through bio-treatment and found that the properties of BRAC were better compared to that of RAC and were comparable to NAC. The technique of preparing the concrete mixture with BRCA through SEMA will extend the utilization of RCA as substitute to NCA in construction. This, in turn will reduce the disposal problems of construction wastes and promote sustainability in the construction.

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Conflicts of interest

The authors have declared no conflict of interest

Data availability statement

The data that support the findings of this study are available within the study and can be collected from the corresponding author upon request.

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