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SELF-HEALING OF MORTAR CRACKS: THE ROLE OF HUMAN PATHOLOGICAL BACTERIA

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ABSTRACT

The characteristics of self-healing related to N-type cement mortars, the addition of human pathological bacteria to enhance mortar durability, and compressive strength thus reducing maintenance cost and manpower after crack initiation. Bacteria have been examined by using two types of biological agents: Proteus mirabilis and E. Coli bacteria. Mortars are formed from a combination of Portland cement and ordinary sand, treated by Proteus mirabilis and E. Coli bacteria with different concentrations. The mechanical properties of the treated mortars are measured and the mortars were characterized by means of Fourier-transform infrared spectroscopy (FTIR), scanning electron microscopy (SEM), and electron energy dispersive spectroscopy (EDX). This study shows that Proteus mirabilis and E. Coli assist the formation of calcium carbonate inside the mortar micro-pores leading to enhancement of the compressive strength and hardness of the mortar specimen. This emphasizes the possibility of activating the self-healing characteristic of the cement mortar and its utilization for cracks treatment and repair.

Keywords: bio-mortar; compressive strength; Pathological Bacteria; urine

ABBREVIATIONS

Abbreviation	Description
BHI	Brain Heart Infusion Broth
DW	Distilled Water
EDX	Electron Energy Dispersive Spectroscopy
EMB	Eosin Methylene Blue
FTIR	Fourier Transform Infrared Spectroscopy Analysis.
NB	Nutrient Broth
OPC	Ordinary Portland Cement
SEM	Scanning Electron Microscopy

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INTRODUCTION

Recently, many studies have focused on concrete and mortar since they are essential building materials. Johnston (1970),Ezeldin (1992), and Rossi (2008) are used in, for example, structures and constructions due to their high compression, and high load-bearing capacity. However, mortars may develop cracks, a serious defect of concrete structure, which leads to its failure and requires immediate treatment and repair Şahmaran (2007),Alahmed (2009), Han (2016). One interesting solution to this problem is now being tested. Researchers consider self-healing agents are suitable to minimize the defect of concrete structure Guo et.al (2018). In self-healing agents, different types of bacteria have been investigated Guo et.al (2018). For example, Fischer et al. (1999), examine the effect of non-pathological bacteria Bacillus Pasteurii on the corrosion protection mortar. They reveal an enhancement of surface protection of sample while adding convenient concentration of calcium and urea to the mixture, due to precipitation CaCO3 Fischer et al. (1999). Hammes et al. (1999-2003), investigate the hydrolyzing of Bacillus pasteurizing bacteria in urea. They show how ammonia and carbon dioxide are formed, and how the latter being calcium carbonate deposited under appropriate micro-environmental conditions. Sporosarcina soli, Bacillus massiliensis, Arthrobacter crystallopoietes, and Lysinibacillus fusiformis (KNUC401, 402,403, and 404), are used as microbiological calcium-carbonate Park et al. (2010). Another important study in this field investigated the effect of different types of bacteria on the compressive strength of mortar Park et al. (2010). They show utilization of Arthrobacter crystallopoietes that leads to improvement of compressive strength, due to the formation of calcium carbonate in its structure. In their study of the effect of different types of bacteria on concrete healing and performance, Sakina et al. (2015) observe positive effects of these agents on the cementitious structures, their durability, and mechanical properties (i.e. improvement of compressive strength). Ganesh & Siddiraju (2016), have studied the effect of bacteria and calcite on the mechanical strength and fracture toughness of the M40 grade concrete. They introduce 3.5-5 % of Bacillus pasteurii and 5-10 % of calcite as partial substituents into the cement mortar samples. They report that compressive strength decreases as calcite contents increase in the concrete structure. Additionally, the bacteria content increased at a constant calcite ratio to help improve the compressive strength. These were attributed to the formation of calcium carbonate inside the concrete structure Ramachandran (2001), Chahal (2012), and Bundur (2015). Meer (2016) & Thakur (2016), have explored the effect of Bacillus Subtilis JC3 on the mechanical properties of M20 grade concrete. They describe an increase in the compressive and tensile strengths of 42% to 63% respectively at 10^5 of bacteria Meer (2016) & Thakur (2016). In addition, they reported several advantages related to, for example, water absorption, durability, and resistance to acidic and chloride agents. These results contribute to the biological agents in the formation of calcium carbonate inside the cracks. However, ammonia existence, as a side effect of reactions, is found to increase the power of hydrogen (ph) and induce precipitations of calcium ions (Ca^{2+}) as nucleation sites due to the negative charge of bacteria cell surface Amirreza (2013) & Guo (2018). Although these ions were assembled outside the cell, they are considered indispensable due to their physiological role in the micro-nutrient of bacteria. Calcium carbonate formation by cell bacteria surface reactions can be recapitulated by Amirreza (2013) & Guo (2018), as demonstrated in (1-4).

$$Ca^{2+} + Cell \longrightarrow Cell - Ca^{2+} \quad (1)$$

$$Cell \xrightarrow[Urea]{Urea} Urease \quad (2)$$

$$\text{Urea} \longrightarrow 2NH_4 + CO2- \quad (3)$$

$$\text{Cell} - Ca + CO_3^{2-} \longrightarrow \text{Cell} + CaCO_3 \downarrow \quad (4)$$

The aim of this study is to investigate the effect of Proteus mirabilis & E.coli bacteria on the mechanical properties, for example, compressive strength, and N-type cement mortars. In this study, a number of characterization measurements are used, for example, scanning electron microscopy (SEM), electron energy dispersive spectroscopy (EDX), and Fourier-transform infrared spectroscopy (FTIR). This paper utilizes the two bacteria at relatively moderate to high concentrations.

Effect of microbiological on concrete

Bacteria can be classified into three groups according to their shape, gram stain, and oxygen demand. Based on oxygen demand bacteria fall into two subgroups; aerobic and anaerobic. It is found that water percolates into cracks when a concrete structure is damaged. As a result, bacteria present in these cracks and receive certain nutrients and water, they start certain processes that generate calcium carbonate, which fills the cracks Muhammad (2016) to Griño (2020). Calcium carbonate is generated by two processes. The first occurs when carbon dioxide reacts with calcium hydroxide Fischer et al. (1999), as in (5).

$$CO_2 + Ca(OH)_2 \longrightarrow CaCO_3 + H_2O \quad (5)$$

Due to the solubility of $Ca(OH)_2$ in water, these molecules can seep into the micro-cracks of the cement mortar. If these cracks are accommodated with bacteria, the latter starts a metabolic activity that generates calcium carbonate, which is an insoluble substance Fischer et al. (1999). Consider the following reaction presented in (6).

$$Ca(C_3H_5O_7)_2 + 7O_2 \longrightarrow CaCO_3 + CO_2 + 5H_2O \quad (6)$$

EXPERIMENTAL WORK

Materials and Method

In this study, ordinary Portland cement (OPC) is used. This cement is procured from the United Cement Company (Al-Mass Basin Co. Ltd.), which complies with the Iraqi specifications of this product Iraqi Specifications … (1984/ No. 5). The physical properties of the cement used in this study are shown in table 1. In table 2, the chemical compositions of cement are presented. In table 3, properties of ordinary sand are given, which meet the Iraqi Specifications … (1984/ 45). In addition, the grading of sand used is displayed in Figure (1), which is used in the cement mortar preparation. Tap water is used for mortar samples preparations, whereas distilled water is prepared for the bacteria (bacteria concentration = CFU/ml 10^5). Calcium hydroxide is procured from (D.B.H Chemicals Ltd., Poole, England). The research process is shown in Figure (2). The two bacteria strains were isolated clinically from urine samples provided by Al Shefa General Hospital in Basrah city. In this method, the urine samples were initially processed and cultured aerobically in blood agar, MacConkey agar, and Eosin Methylene Blue (EMB) agar plates for 24 h at 37 °C. According to Bergey's manual of systematic bacteriology, the phenotypic colony, microscopic cell identification through Gram staining, and biochemical tests were performed. In order to classify the isolated bacteria according to the Gram-negative identification protocol, the conventional biochemical tests and Vitek®2 automated systems

were also performed J. M. Miller (2018). Next to the diagnosis process, all the bacterial strains were activated in nutrient broth (NB), brain heart infusion (BHI) broth as well as in distilled water (DW), as shown in plates 1 and 2. N-type cement mortars were prepared with mixing ratios of 1:1:6 and w/c = 0.5. Dry heterogeneous materials (i.e. cement and sand) were first admixed for 2 minutes. Later, the activated bacterial strains were added to the mortars and mixed for 10 minutes. Table 4 shows these processes in detail. Finally, water is added to the mixture and mixed with hand for 10 – 15 minutes before molding of the concrete samples. The same procedure is adopted for each concentration for the bacteria used. The curing time was set to 28 days.

RESULTS AND DISCUSSION

Three cylindrical samples were used for the compressive strength test. In addition, a hydraulic machine (0.5 mm/min) was used. The compressive strength test results of the cement mortars, cured for 28 days, are shown in Figure 3. As the number of bacteria increases in the cement mortars, compressive strength is found to be increased consistently. This behavior is of considerable value for samples containing 5 ml of Proteus mirabilis activated in nutrient broth. Compressive strength increases at 97.7 %. After conducting the destructive test (Compressive strength), visible cracks are generated in cement mortar. As a result, the bacteria activated to contribute to the formation of calcium carbonate inside the microspores to fill the crack formed, which occurred within approximately (10 days) after the mechanical test. These results agree with the results of research that used bacteria from soil Park (2010), Chahal (2012), and Thakur (2016). In Figure 4, the results of hardness test (ASTM D2240) Koch T., the loading force of shore hardness D: ($0\ N \leq F \leq 44.5\ N$) with spherical cap ($0\ mm \leq h \leq 2.5\ mm$), with range value (0 – 100) Koch T., are described. Hardness was found to increase with the increase of bacteria concentration and this behavior is highly remarked in cement mortars treated with 15 ml of Escherichia coli activated in the brain heart, 5 ml of Proteus mirabilis activated in nutrient broth and 25 ml of Escherichia coli activated in nutrient broth. These results are consistent with compressive strength values. The Fourier transforms infrared spectroscopy is performed in the range 500-4000 cm^{-1} while using FTIR spectrometer from Shimadzu (Shimadzu Japan) A. AL-Zubaid (2017). FTIR results are shown in Figures 5, 6, and 7 for the BHI, Nutrient broth E. Coli bacteria, and *Proteus* bacteria. Broadbands of calcium hydroxide occur in the range 3631.96- 3410.15 cm^{-1} while calcium carbonate gives spectroscopic peaks in the range 1440- 1489 cm^{-1}. These peaks are directly related to the effect of bacteria in the cement mortars. This is what is provided with cement mortar its high hardness. Research sustained by scanning electron microscope is a useful technique in studying texture properties of solid objects such as additives, micro-cracks, and new phases. Figure 8 (a) and (c) show the 5 ml Nutrient broth Proteus contained cement mortars under scanning electron microscopes. The corresponding energy dispersive X-ray spectroscopy results are shown in Figures 8 (b) and (d). The scanning electron micrographs of mortar (with and without additives) are shown in Figure (9), that appeared addition of Proteus mirabilis activated in nutrient broth in which a decrease of micro-pores in the cement mortar was a consequence. This is accompanied by an increase in the calcium carbonate in the cement mortar, as indicated by the EDS results. As can be seen, the increase of $CaCO_3$ in the cement mortar improves the mechanical properties.

CONCLUSIONS

In light of this evidence, it is clear that the results of this study support studies using types of soil bacteria such as:

1. Bacillus Pasteurii, and Arthrobacter crystallopoietes, which shows its positive effect on compressive strength. In contrast, this study reveals that the utilization of two types of human pathological bacteria (Escherichia coli and Proteus mirabilis), as biological agents in cement mortars, is accomplished successfully.
2. This occurs while contributing to the self-healing of micro-pores and enhancement of compressive strength and hardness of the cured mortars. The compressive strength increases by 97.7 % for the cement mortars treated with 5 ml of Proteus mirabilis activated in nutrient broth, by 64.5 % for mortars treated with 15 ml of Escherichia coli activated in brain heart broth, and by 51.7 % for those specimens treated with 25 ml of E.coli activated in Nutrient broth.
3. FTIR, SEM, and EDS measurements show that these biological agents produce calcium carbonate in the treated specimens, which are fundamental in self-healing characteristics that are promising procedures for mortars cracks treatment and repair.

Table 1. Physical properties of OPC.

Type of Test	**Results**	**Limit of IQS.No.5**
Specific Surface [m2 /kg]	376	>230
Setting Time (Vicate method) -Initial setting [hrs:min] -Final setting [hrs:min]	 2:05 4:00	 ≥ 45 [min] < 10 [hrs]
Compressive Strength of Mortar -3 Days -7 Days	 20 [MPa] 25 [MPa]	 ≥ 15 ≥ 23
Autoclave (Soundness)	0.12	≤ 0.8

Table 2. Chemical composition of OPC.

Oxide composition	**Abbreviation**	**%by weight**	**Limits of IQS No. 5/1984**
Lime	CaO	66.11	-
Silica	SiO_2	21.93	-
Alumina	Al_2O_3	4.98	-
Iron oxide	Fe_2O_3	3.10	-
Sulphate	SO_3	2.25	2.8% ≥
Magnesia	MgO	2.0	5%≥
Loss of Ignition	L.O.I.	2.39	4% ≥
Lime saturation factor	L.S.F.	0.93	0.66-1.02
Insoluble residue	I.R.	1.29	1.5 ≥
Main compound (Bouge eq.)		**By weight of cement**	
Tricalcium silicate	C_3S	58.16%	-
Diacalcium silicate	C_2S	18.997	-
Tricalcium aluminate	C_3A	7.95	-
Tetracalcium aluminoferrite	C_4AF	9.43	-

Table 3. Physical Properties of Sand.

Physical Properties	Results	Limit of IQS. No. 45/1984
Specific Gravity	2.6 %	-
Sulphate Content (SO_3)	0.343 %	Max.= 0.5%
Water Absorption	2%	-
Clays and Fine Minerals	4%	Max.= 5%
Fineness Modulus	2.8	-

Table 4. Bacteria concentration in cement mortar

No.	Human Bacteria Type	Concentration (ml)
1	Control specimen	10
2	Proteus mirabilis activated in brain-heart infusion broth	10
3	Proteus mirabilis activated in nutrient broth	10
4	Escherichia coli activated in brain-heart	10
5	Escherichia coli activated in nutrient broth	10
6	Escherichia coli from distilled water	10
7	Proteus mirabilis from distilled water	10
8	Proteus mirabilis activated in brain-heart infusion broth	20
9	Escherichia coli activated in brain-heart	20
10	Proteus mirabilis activated in brain-heart infusion broth	15
11	Escherichia coli activated in brain-heart	15
12	Escherichia coli activated in nutrient broth	25
13	Proteus mirabilis activated in nutrient broth	5

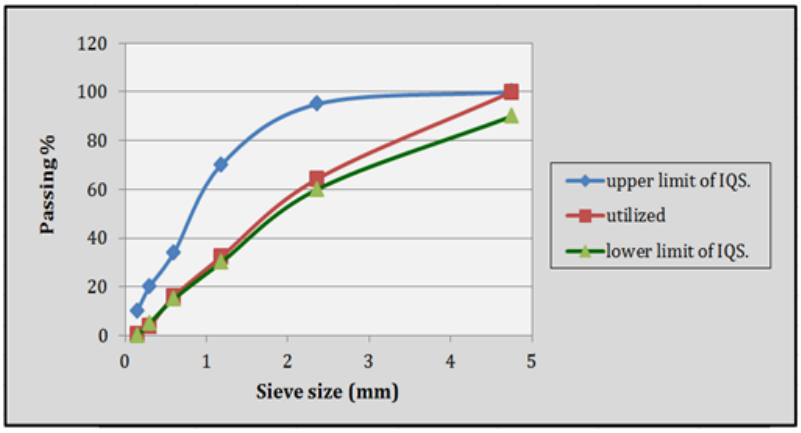


Fig.1. Grading of natural sand **Iraqi Specifications Measurement…(1984/ 45)**.

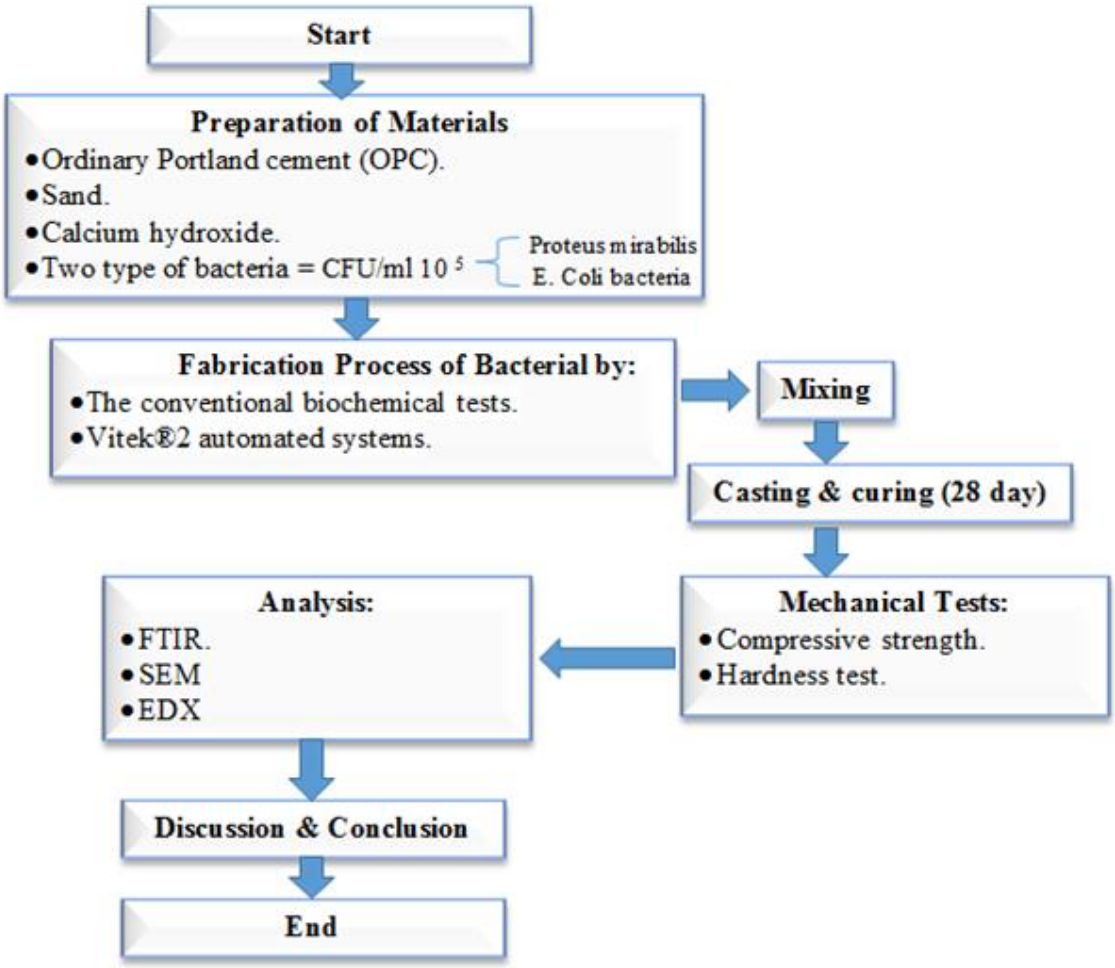


Fig. 2. Flowchart of research methodology

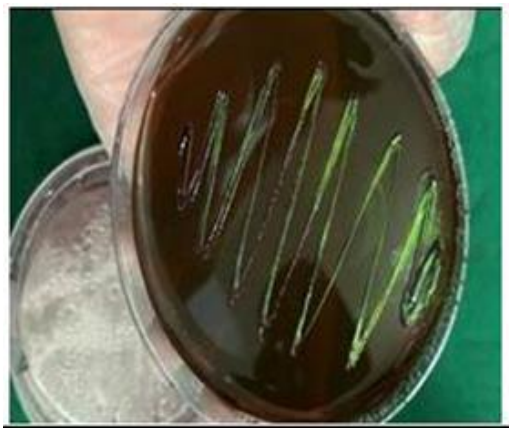


Plate 1. E.coli on EMB agar.

Plate 2. E.coli activated in BHI broth.

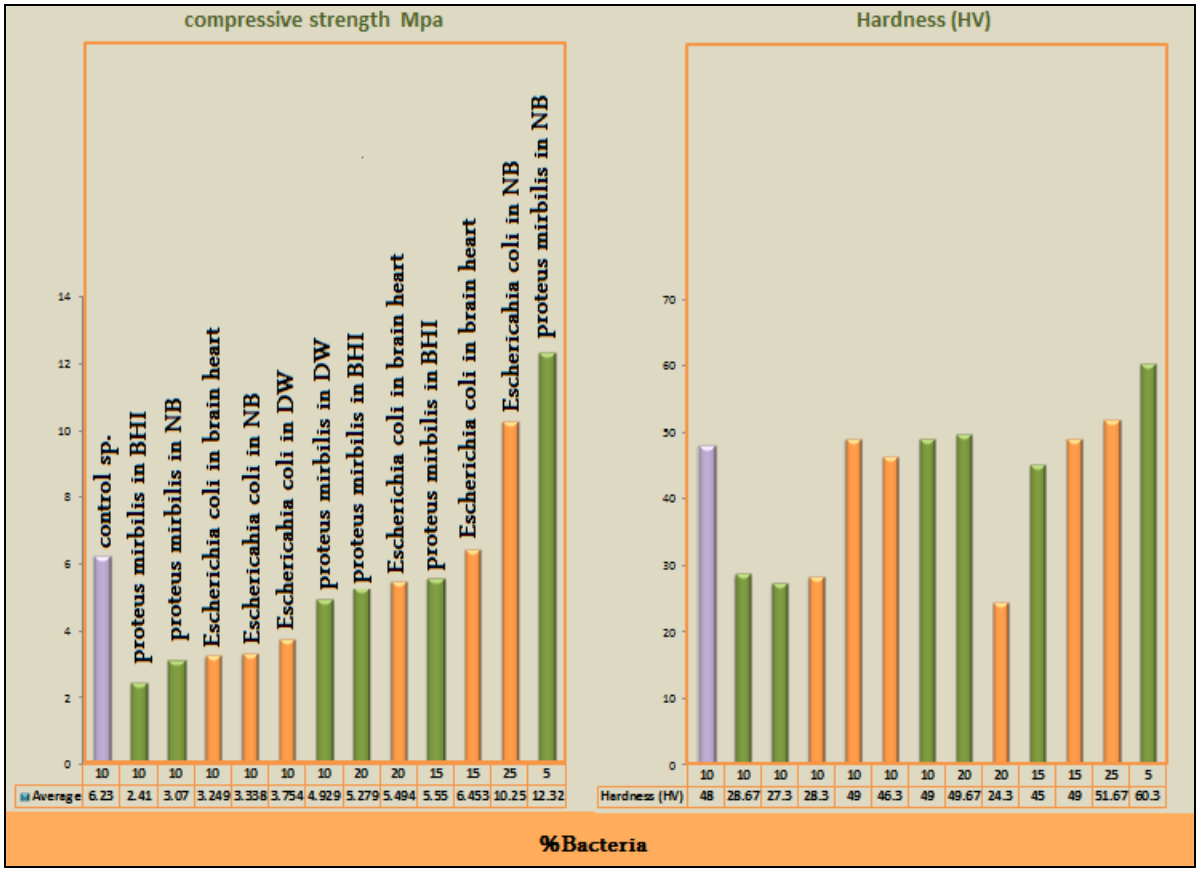


Fig. 3. Compressive strength for bio-(N-type) mortar. Fig. 4. Hardness test for bio-(N-type) mortar.

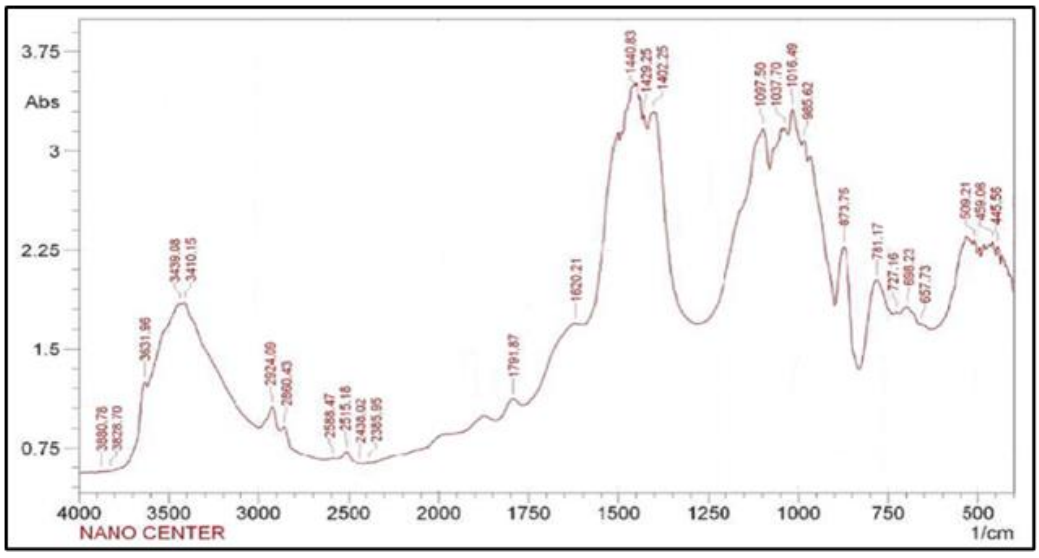


Fig. 5. FTIR spectrum of cement mortar with 5ml of BHI E.Coli bacteria

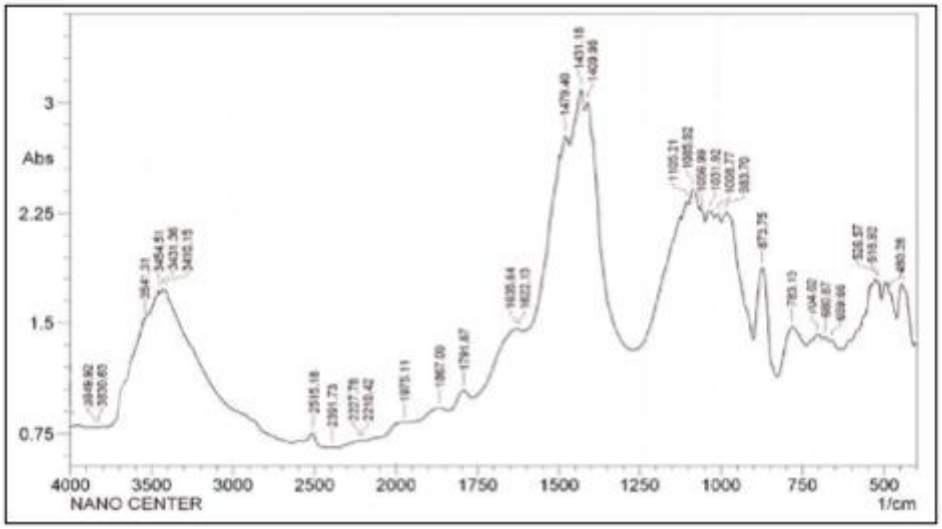


Fig. 6. FTIR test for bio-mortar with (25 ml) of Nutrient broth E.coli bacteria.

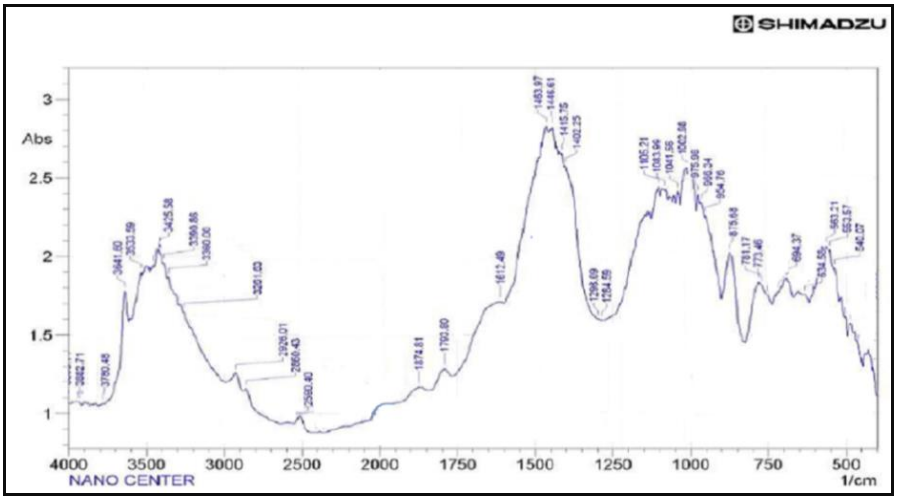
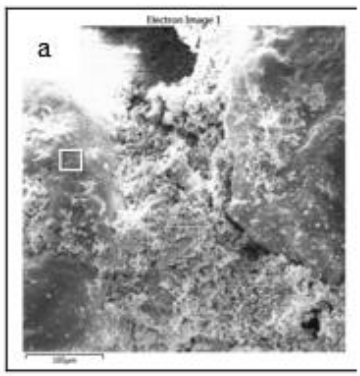
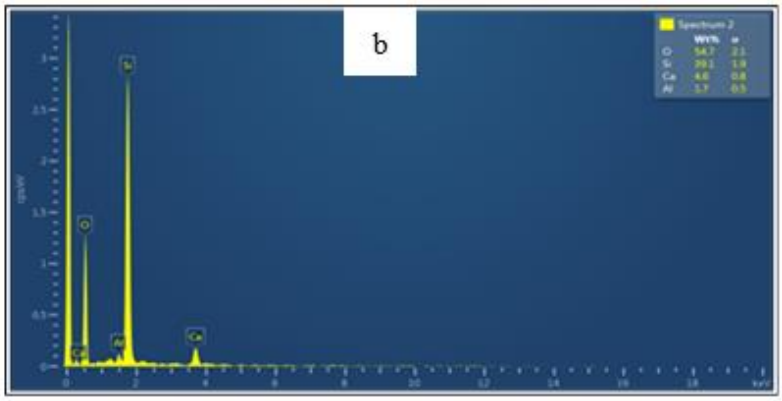


Fig.7. FTIR test for bio-mortar with (25ml) of Nutrient broth Proteus bacteria.





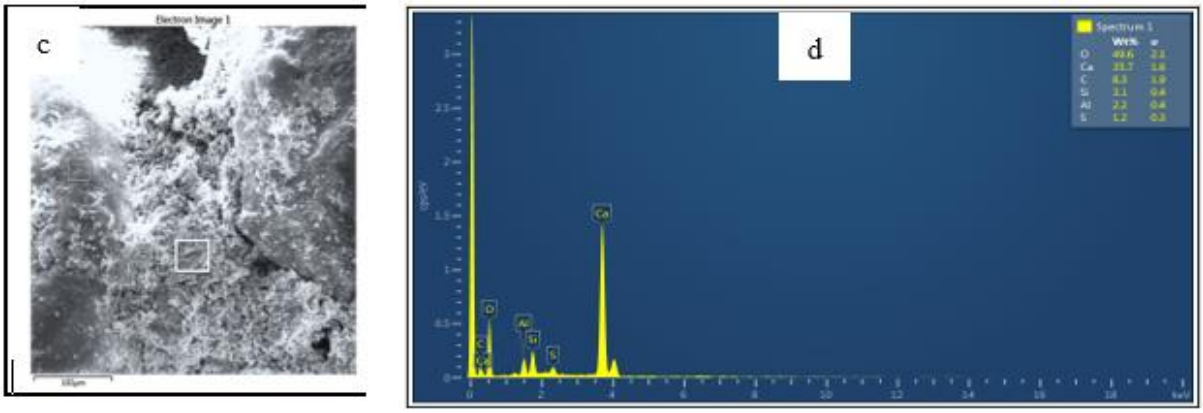


Fig.8. (a, c) SEM for bio-mortar with (5ml) Nutrient broth Proteus. (b,d) EDX for bio- mortar with (5ml) Nutrient broth Proteus.

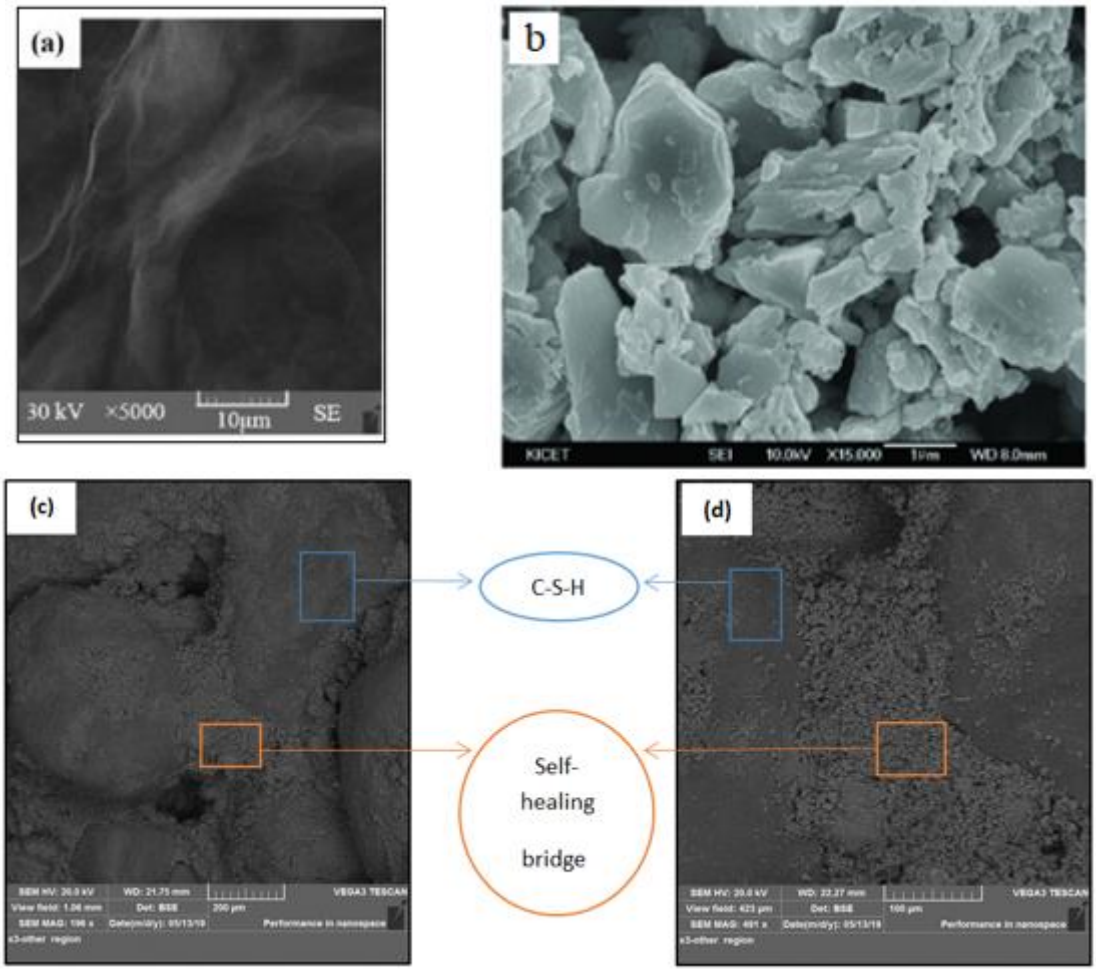


Fig. 9. (a) SEM of base mortar, (b) SEM of cement powder, (c) & (d) SEM for bio-mortar with (5ml) Nutrient broth Proteus.

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