Algae as a part of microorganisms involved in biocorrosion of cement composites with total replacement of natural aggregates by photovoltaic glass

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Abstract. Algae of the Pleurococcus, Trentepohlia and Stichococcus genera were selected for the experiment on the cement composites with total replacement of natural aggregates by recycled photovoltaic glass. The growth of the algae was monitored on cement beams with dimension 40 x 40 x 8 mm (length x width x height) and on the cement crumbling in various proportion. In the case of cement composites with recycled glass from photovoltaic panels, intensive growth was observed in the Trentepohlia genus from the Chlorophyceae class, while in the case of cement crumbling with recycled photovoltaic glass, the most obvious growth in biomass was recorded in the Pleurococcus genus; the Stichococcus and Trentepohlia genera showed minimal or zero growth. This minimal growth is mainly influenced by the pH value and the fact that biocorrosion is accompanied by the effect of symbiotizing microbiota, which mutually support each other, not just one species.

1. Introduction
Biocorrosion is the common name for the disruption of the properties of materials by organisms, in most cases microorganisms or lower plants, and their metabolic activity. Biocorrosion takes place not only in building materials, but it can also be observed on archived books, pipeline structures, statues and monuments. This negative phenomenon results in the decrease in the quality of individual composites, the overall quality of building structures, as well as their appearance [1]. Biodeteriogen, or an organism involved in biocorrosion, causes undesirable changes in cement composites with its metabolites. The main organisms involved in biocorrosion are especially microorganisms - cyanobacteria, algae, micromycetes and bacteria, but they can also be lower plants - lichens or bryophytes [2,3]. If the biodeteriogen damages composites with its metabolic products, it is dissimilation damage, while if the composite represents food for the biodeteriogen, it is assimilation damage [4]. Like any other processes, the biocorrosion process is affected by several factors. Due to the involvement of microorganisms in this process, the most important factors include nutrients, temperature, humidity and pH. Nutrients for microorganisms are represented by inorganic and organic substances present in cement composites [5,6], humidity is essential for the existence and occurrence of microbial consortia on the surface of buildings, temperature affects the growth rate of biodeteriogens and pH value is associated with the present microbiota activity and its nutrient intake from the environment [7-9].

The average lifespan of photovoltaic panels is estimated at 25 years [10, 11] which is why it is necessary to look for a solution focused on what to do with the resulting waste. Directive 2012/19 / EU of the European Parliament and of the Council on waste electrical and electronic equipment from 15 August 2018 [12] stipulates that at least 85% of materials from photovoltaic panels must be used and 80% of materials must be ready for re-use and recycled. It is estimated that photovoltaic panel waste, which
consists mainly of glass, could reach 78 million tonnes worldwide by 2050 [13]. One of the proposed solution suggesting how this glass could be handled is to incorporate it into a cement matrix [14,15].

2. Methods
This experiment was focused on the effect of microscopic algae on cement composites with total replacement of natural aggregate by recycled glass originating from photovoltaic panels at the end of the life cycle. The growth of selected species of algae was monitored on cement beams and cement crumbling.

2.1. Algae
Algae of the Pleurococcus and Trentepohlia and Stichococcus genera were used in the experiment. Pleurococcus is a genus of green algae from the Chlorophyceae class having a strong cell wall that protects them against high water losses. They have spherical shape and can be found both individually and in bundles, where they form a thick mucilaginous layer. They are also part of the biofilm. In the process of biocorrosion, their metabolic processes produce organic acids which dissolve the individual components of building stone [16,17].

The genus of green algae Trentepohlia also belongs to the class of Chlorophyceae. This cosmopolitan algae form close symbiosis with lichens, which are also involved in biocorrosion. Their metabolites release carbon dioxide, which dissolves the carbonate components contained in cement composites in the form of acid carbonates [1,18,19].

The genus of Stichococcus is a photosynthetic green alga with a polysaccharide cell wall living aerophytically and in symbiosis with lichens. This algae causes ugly patina on the surfaces and also grows in the pores, while its growth penetrates building stone and causes its instability [18,20].

2.2. Preparation of cement composites with total replacement of natural aggregate
The cement composites were prepared on the basis of recycled glass from solar panels. The input components for the production of cement composites were as follows: photovoltaic glass as a total replacement of natural aggregate which consisted of 4 fractions (0/0.5 mm; 0.5/1 mm; 1/4 mm; 4/10 mm), Portland cement EN 197-1 - CEM I 52.5 R and mixing water from the water supply system. The R0 recipe was designed as a comparative one and contained total of natural aggregate. Based on the determined optimal curves of recycled glass grain size, 5 new recipes (R1-R5) for the production of concrete mixture were designed. The designed recipes contained different ratio of the individual fractions. Only two recipes contained all fractions (R3 and R5). The remaining three recipes (R1, R2 and R4) did not contain 1/4 mm fraction photovoltaic glass recylcate. The mixture was worked into beam moulds with dimension 40 x 40 x 160 mm (W x H x L), the mixture was left in the mould until the next day and then the mould was removed. The resulting beams were stored in an aqueous environment at 20 °C for 28 days, and then they were removed and dried to a constant weight. The treatment procedure of the test specimens was as follows:

- test specimens with dimension 40x40x8 mm were used for the 1st experiment, they were treated with ethanol to prevent drying of the medium with microorganisms, due to length of the experiment.
- crumbling (prepared by crushing hardened composites) was used for the 2nd experiment as a substitute for 10%, 20% and 40% of agar.

2.3. Cultivation medium
Algae Cultura Agar medium (HiMedia Laboratories, Mumbai, India) was used for algae cultivation with these main components: sodium nitrate, dipotassium phosphate, magnesium sulphate, ammonium, calcium and ferric chloride, agar and distilled water, pH 7±0.2. The medium was prepared in Erlenmeyer flasks and sterilized in a Sterimat Plus autoclave at 121 °C.
2.4. The course of experiment

The cut test specimens were prepared according to the recipes R0 - R5 and were placed in petri dishes (diameter 90 mm) and embedded with sterile cultivation medium to the level of the samples. Subsequently, after the medium had solidified, microscopic algae were inoculated and the petri dish prepared in this way was secured with a parafilm to prevent water leakage and a possible contamination from the environment. In the case of cement crumbling, a petri dish with 10%, 20% and 40% cement crumbling was prepared, and it was subsequently supplemented to 100% using the medium. After the medium had solidified, microscopic algae were inoculated and the petri dish prepared in this way was secured with a parafilm. The experiment with the prepared cement crumbling took place on samples prepared according to recipes R1-R5.

All the prepared samples were placed in a laboratory with sufficient sunlight and the growth of algae was checked at intervals during 28 days. It was subsequently evaluated according to the ČSN 72 4310 standard [21].

3. Results and discussion

The degree of algae growth was evaluated according to the standard [21], which determines the degree of fungal growth. There is currently no relevant standard for assessing algae growth.

0 - fungi do not grow; 1 - growth is negligible; 2 - growth is gradual (up to 25%); 3 - growth is intensive (up to 50%); 4 - growth is very intensive (up to 75%); 5 - growth is complete (100%).

Table 1 shows the most intensive growth in the genus of Trentepohlia, however it did not exceed the value of 3 - intensive growth up to 50% of the surface, namely in recipes R1 and R2. The same growth trend was observed in recipes R3 and R4: 2 - gradual, up to 25% of the surface throughout the entire measurement period. The lowest growth activity was observed in the R0 recipe. The genera of Pleurococcus and Stichococcus showed only increase to the value of 1 - negligible in all recipes R0 - R5.

| Algae species | Pleurococcus | Stichococcus | Trentepohlia |
|---------------|--------------|---------------|--------------|
| Days          | 7 14 21 28   | 7 14 21 28   | 7 14 21 28   |
| Recipe        | Pleurococcus | Stichococcus | Trentepohlia |
|----------------|-------------|---------------|--------------|
| R0             | 1 1 1 1     | 0 1 1 1      | 1 1 1 1      |
| R1             | 1 1 1 1     | 1 1 1 1      | 1 3 3 3      |
| R2             | 1 1 1 1     | 1 1 1 1      | 2 3 3 3      |
| R3             | 1 1 1 1     | 0 0 1 1      | 2 2 2 2      |
| R4             | 1 1 1 1     | 0 1 1 1      | 2 2 2 2      |
| R5             | 0 0 1 1     | 1 1 1 1      | 2 2 3 3      |

Tables 2-4 show the growth intensity of algae biomass in the ratios of 1:9, 2:8 and 4:6 of the crumbling to medium sample. Overall, Pleurococcus was the most successful in all three ratios, however, the growth activity did not exceed the level of 3 - intensive growth, up to 50% of the surface; the species of Stichococcus showed an absolute absence of growth in all ratios and in the species of Trentepohlia the growth was insignificant, mainly at 10% of the crumbling to medium ratio. In case of the highest share of crumbling - 40%, the growth was the lowest overall, which is attributed to a very small amount of nutrients present in the medium. The composition of recipes R1-R5 had no significant effect on the support or inhibition of the growth of microscopic algae.

| Algae species | Pleurococcus | Stichococcus | Trentepohlia |
|---------------|--------------|---------------|--------------|
| Days          | 7 14 21 28   | 7 14 21 28   | 7 14 21 28   |
Recipe  

Table 3 Biocorrosion rate on cement crumbling samples with total replacement of natural aggregate with photovoltaic glass in 20% representation of the sample during 7, 14, 21 and 28 days. 

| Algae species | Pleurococcus | Stichococcus | Trentepohlia |
|---------------|--------------|--------------|-------------|
| Days          | 7 14 21 28   | 7 14 21 28   | 7 14 21 28  |
| Recipe        |              |              |             |
| R1            | 1 1 1 1      | 0 0 0 0      | 1 1 1 1     |
| R2            | 1 2 2 2      | 0 0 0 0      | 1 1 1 1     |
| R3            | 2 2 2 2      | 0 0 0 1      | 1 1 1 1     |
| R4            | 2 2 3 3      | 0 1 1 1      | 0 0 1 1     |
| R5            | 2 2 2 2      | 0 0 0 0      | 0 0 1 1     |

Table 4 Biocorrosion rate on cement crumbling samples with total replacement of natural aggregate with photovoltaic glass in 40% representation of the sample during 7, 14, 21 and 28 days. 

| Algae species | Pleurococcus | Stichococcus | Trentepohlia |
|---------------|--------------|--------------|-------------|
| Days          | 7 14 21 28   | 7 14 21 28   | 7 14 21 28  |
| Recipe        |              |              |             |
| R1            | 0 0 0 0      | 0 0 0 0      | 0 0 0 0     |
| R2            | 0 1 1 1      | 0 0 0 0      | 0 0 0 0     |
| R3            | 0 0 0 0      | 0 0 0 0      | 0 0 0 0     |
| R4            | 0 1 1 1      | 0 0 0 0      | 0 0 0 0     |
| R5            | 0 0 0 0      | 0 0 0 0      | 0 0 0 0     |

Figure 1 shows samples of beams (recipes R1-R5) with growing biomass of Stichococcus algae after 28 days of growth evaluation and Figure 2 shows individual colonies of these algae under magnification. 

Figure 1. Samples of beams inoculated with Stichococcus algae after 28 days of growth. 

Figure 2. Detailed view of Stichococcus algae colonies after 76x magnification.
4. Conclusion
This experiment was focused on the biocorrosion of cement composites - beams and crumbling, with total replacement of natural aggregate with photovoltaic glass in four different grain fractions. Many microorganisms are involved in the process of biocorrosion, and they are influenced by the surrounding environment factors as well as other organisms present [22]. However, three species of microscopic algae - *Pleurococcus*, *Stichococcus* and *Trentepohlia* - were used in this experiment because they were identified as part of the biofilm involved in biocorrosion [23]. Based on the presented results, we can draw the following conclusions:

- the growth of microscopic algae was quantitatively small;
- low abundance was caused by the absence of symbiotizing microbiots from the point of view of growth support of selected algae representatives;
- growth quality was also affected by the high pH value of the samples (pH>11);
- weak growth of the used algae was also affected by the high level of recycled glass present in the samples, mainly in the experiment with crumbling, which affected the content of nutrients available for algae nutrition.

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