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Testing of algae colonization growth risk on building materials

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Abstract. Due to biological colonization, properties of building materials change. Green algae represent one group of the entire complex of biodeteriogens that interact with each other. For the purpose of testing algae growth risk on concrete with varied composition, a growth resistance testing methodology was developed based on ČSN EN 15458 (672032) *Paints and varnishes - Laboratory method for testing the efficacy of film preservatives in a coating against algae*. The modified method was employed to test ten samples of concrete with different composition, i.e. two types of cement and two types of classical fly ash were incorporated in the samples. The green algae growth was influenced by both the type of cement and the type of fly ash used. The results show that any addition of classic fly ash increases the risk of green algae colonization on the surface of concrete.

1. Introduction

Green algae and cyanobacteria require light, air humidity, and minerals to grow. They grow very well in places where water is retained (cornices, window sills, balconies, uneven zones of masonry, plinths), especially in the northern and north-eastern parts of buildings, including roofs. Algae also occur more frequently on spots where trees and bushes grow close to buildings and shade for a significant part of the day. Under favourable humidity conditions, extensive layers of different consistencies, thicknesses and colours appear [1]. Research shows that the first biofilm emerging on a surface consists of various types of bacteria followed by algae. Soon after algae, microscopic fibrous fungi colonize the biofilm, and later mosses and lichens are emerging to prepare the substrate for higher plants growth. On building stones, green algae are most common, namely the genera *Gloeocystis, Tetracystis, Cystomonas, Muriella, Chlorella, Stichococcus, Scenedesmus, Klebshormium, Apatococcus, Trebauxia*). Another large, frequently observed group consists of cyanobacteria (*Aphanothece, Lyngbya, Leptolyngbya, Nostoc, Synechococcus, Plectonema, Phormidium, Chroococcus, Myxosarcina, Pleurocapsa, Scytonema, Gloeocapsa, Apatococcus, Stichococcus, Phormidium*) [2]. Algae are able to grow in very extreme conditions, i.e. not only survive but even actively metabolize. Many species are known for their ability to survive for a long time at extreme temperatures and many also tolerate drying as well as high or low pH, strong gamma and UV radiation. On stone and building facades, algae can remain inactive for several years and return to biofilm under appropriate conditions. Their durability is due to presence of protective outer coverings which also serves as a reservoir of water that is held there by molecular forces. The cyanobacteria and algae shells also contribute to thickness and stability of the biofilm crusts they create on the building.

The surface of the building stone is mainly affected by algae metabolites. Algae synthesize polysaccharides, amino acids, vitamins, organic acids such as citric, glutamic, glycolic, oxalic, uronic and others, nitrogen bases, dyes and other substances. These products can be utilized by associated bacteria and moulds that produce more acids, especially formic acid, acetic acid, oxalacetic acid, malic

acid, succinic acid, glyoxalic acid, gluconic acid, glucuronic acid and oxalic acid. This leads to acidolysis of the building stone and extensive formation of efflorescences containing increased concentrations of Ca^{2+}, K^{+}, Na^{+}, Mn^{2+}. The acid anions react with the cations of the stone to form water-soluble salts, which hydrate, increase the water content in the building stone pores and subsequently cause cation release from the building stone, so-called chelation. This leads to increased porosity and gradual decay of the building stone surface [2]. Algae also act mechanically, growing into small slits and expanding them. Some types of algae actively penetrate the building stone, create microgaps of various shapes and their secretions dissolve and leach carbonates present in the building stone. Fibrous green algae create slime-like coatings and crusts consisting of their fibres and plaster particles glued together. During the year, alternate drying and moistening of this biofilm occurs, as well as changes during freeze-thaw cycles, which together leads to removal/decorticating of the algae substrate. Thus, the top layer of the plaster or stone detaches from the deeper layers. The surface distortion process then proceeds in the following order: algae colonize humid cracks in stone sized from 0.1 to several millimetres in diameter, grow in dependence on the presence of water, carbonates around the cells precipitate in a reversible process $Ca(HCO_3)_2 \leftrightarrow CaCO_3 + H_2O + CO_2$, gradually cracks open by continuous carbonate precipitation and algae expansion with water inhibition, dust particles penetrate the cracks, algae die and the crack is colonized by heterotrophic bacteria and fungi, possibly insects, mites and spiders. The process leads to increased pressure on crack walls by newly produced organic components and to the removal of surface layer [3].

The aim of this research was to modify and optimize the method for evaluation of resistance of concrete samples to green algae. Subsequently, the modified method was used to test the effect of concrete composition, especially the addition of fly ash with a proven negative effect on the growth of water algae.

2. Green algae resistance testing methodology

The proposed methodology is based on ČSN EN 15458 (672032) Paints and varnishes - Laboratory method for testing the efficacy of film preservatives in coating against algae [4]. It is the only standard dealing with algae influence on building materials.

Testing bodies sized 75 × 75 × 10 mm were prepared. A standard formulation of fine-grained concrete was proposed for the test samples for the basic verification of the proposed methodology for testing the resistance to algae and moulds. Standard sands CEN I, CEN II and CEN III were chosen as fillers to eliminate impurities in concrete that could distort the results of the evaluation. Equal amounts of each sand were added into all mixtures. Two types of binders were used: CEM II S-M (S-LL) 32.5R cement and CEM I 42.5R cement. Therefore, two types of concrete mixtures (cement + sand mixture + water) were prepared and used as standards – standard 1 ST (cement CEM I) and standard 2 ST (cement CEM II). In the proposed P25 and P50 formulations, 25 and 50% of standard sand was replaced with fly ash selected from many samples obtained from different producers across the Czech Republic. Based on the results of previous analyses of the ashes, two types of classical fly ash were selected. These two ashes showed ecologically unfavourable properties, i.e. excessive levels of hazardous substances concentration in the leachate and unsatisfactory results of ecotoxicity testing. According to the authors' theory, these unfavourable properties should inhibit the algae growth on the concrete mixture surface. Ashes are indicated in the formulations as P1 and P2. The testing bodies were cured for 28 days and sterilized by UV light (30 min) prior to the test itself. Two bodies of each formulation were tested.

For the inoculation of testing bodies, an algal suspension of two algae species was used: *Nostoc commune* and *Stichococcus bacillaris*. The algae cultures were purchased from the collection of autotrophic organisms CCALA Třeboň (*492 Stichococcus bacillaris Naegeli, 118 Nostoc commune Vaucher*). The strains were grown and inoculation suspensions were prepared according to the standard (density of 10^6 CFU/ml of the suspension). The Bold-Basal/Bristol Medium (BBM medium) was used both for growing strains and for testing. Composition of stock solutions for the BBM medium preparation is listed in Table 1.

Table 1. Stock solutions.

Number of solution	Dissolved chemicals	Concentration
1.	$NaNO_3$	25 g/1000 ml
2.	$CaCl_2.2H_2O$ (or $CaCl_2.6H_2O$)	5 g/1000 ml (or 3.72 g)
3.	$MgSO_4.7H_2O$	7.5 g/1000 ml
4.	K_2HPO_4	7.5 g/1000 ml
5.	NaCl	2.5 g/1000 ml
6.	KH_2PO_4	17.5 g/1000 ml
7.	EDTA+KOH	5 g disodium EDTA + 3.1 g KOH /100 ml
8.	$FeSO_4.7H_2O$	0.498 g + H_2SO_4…..0.1 ml /100 ml
9.	H_3BO_3	1.142 g/100 ml
10.	Microelements solution – 1000 ml	
	$ZnSO_4.7H_2O$	8.82 g
	$MnCl_2.4H_2O$	1.44 g
	MoO_3 (or $Na_2MoO_4.2H_2O$)	0.71g (or 0,242 g)
	$CuSO_4.5H_2O$	1.57 g
	$Co(NO_3)_2.6H_2O$	0.49 g

The BBM medium itself is prepared by mixing 10 ml of each of solutions 1–6 and 1 ml of solutions No. 7–10 with deionized water in a 1000 ml volumetric flask. For the test, a solid substrate is prepared from the nutrient BBM medium by addition of 1.5% of agar. This suspension is autoclaved and afterwards poured into Petri dishes to form circa 0.5 cm thick layer. The testing concrete bodies were placed on the surface of the layer, one into each dish. Both the body and the surface of the substrate were covered with a thin layer of mixed algal culture at a density of 10^6 CFU/ml, as 10 ml of culture suspension was poured onto the test specimen and flowed down onto the substrate. Two cultivation controls were prepared. In the first case, the algal suspension inoculated only the agar, in the latter case a square of filter paper of the sample size was placed on the agar substrate.

Incubation was carried out at 23 ± 2°C, 1000 ± 200 lx, 16 h light/8 h dark cycle. The test lasted for 35 days. Since the 20th day of the test, the samples and the agar were sprayed with BBM nutrient medium to avoid drying out.

After 14, 21, 28 days and the end of the test, the algae growth on the test bodies was visually evaluated, both macroscopically and using a stereomicroscope. Gradual growth of culture in control dishes was evaluated and also, possible contamination with other types of microorganisms was observed. The final evaluation of the test results was carried out according to the standard by means of degrees 0–3 (0 no growth, 1 low growth, 2 medium growth, 3 heavy growth). For the comparison of the samples, the + and - signs were used for specification.

3. Results

In Figure 1, 2, 3, 4 a photodocumentation of sample 2-P2-25 during the test is listed. The algae growth was macroscopically observed after 14 days of duration, while the effect of cement mixture formulation on growth was observable after 21 days.

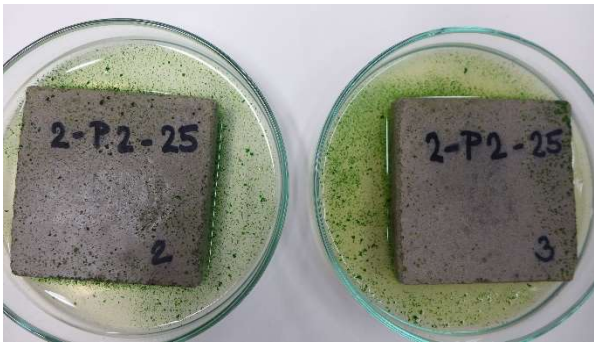

Figure 1. Development of algae growth during the test – sample 2-P2-25 14 days of cultivation.

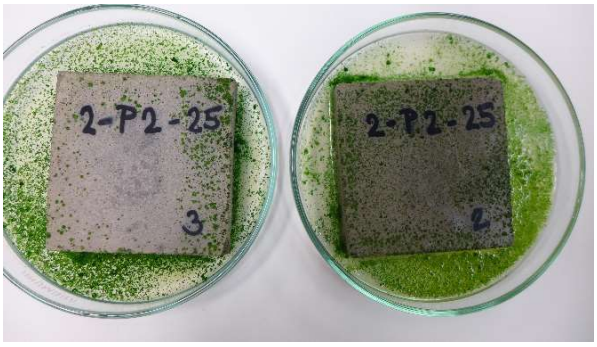

Figure 2. Development of algae growth during the test – sample 2-P2-25 21 days of cultivation.

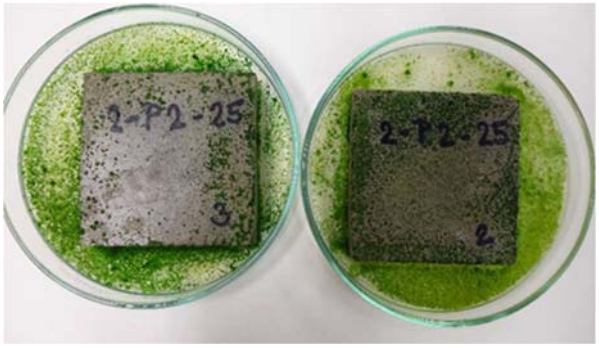

Figure 3. Development of algae growth during the test – sample 2-P2-25 28 days of cultivation.

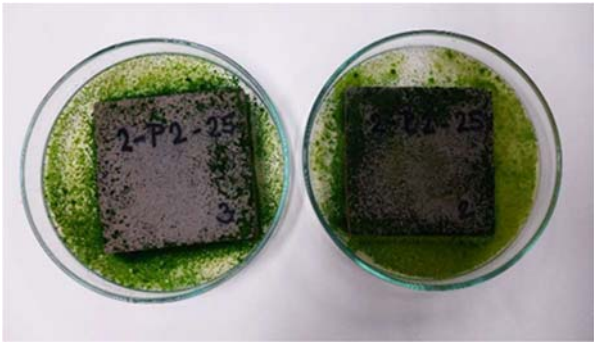

Figure 4. Development of algae growth during the test – sample 2-P2-25 35 days of cultivation.

In Figure 5, 6 condition of selected samples at the end of the test is listed. No undesirable contamination of the samples was detected.

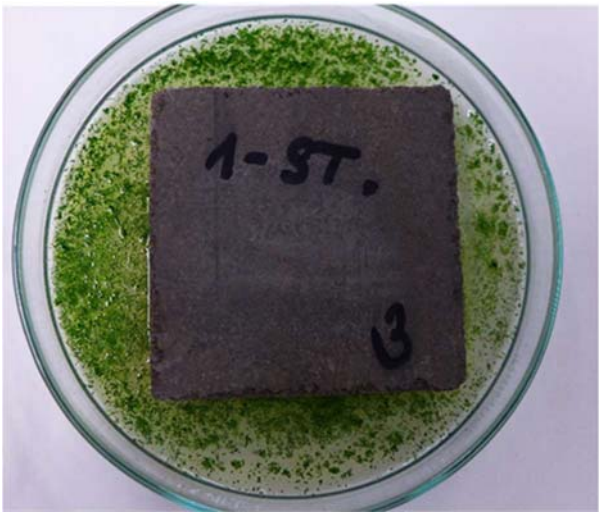

Figure 5. Algae growth on the last day of the test – sample 1 ST.

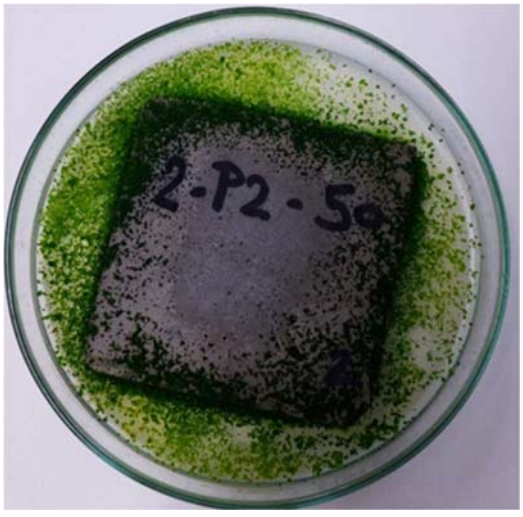

Figure 6. Algae growth on the last day of the the test – sample 2-P2-50.

The last day of the test, algae growth was evaluated using degrees 0–3 (0 no growth, 1 low growth, 2 medium growth, 3 heavy growth). For the comparison of the samples, the + and - signs were used for specification and the results were summarized in Table 2.

Table 2. Algae growth evaluation at the end of the test.

Sample	Degree of growth
1 ST	0
2 ST	2
1-P1-25	2-
1-P1-50	2+
1-P2-25	1
1-P2-50	1+
2-P1-25	3
2-P1-50	3+
2-P2-25	2+
2-P2-50	2-3

The final results fully correspond with the concrete mixtures formulation. Both the type of cement and the type of fly ash influence the algae growth on the surface of the samples. Among the four studied concrete ingredients, the highest inhibition effect exhibited cement CEM I 42.5R and classical fly ash P1. Where these two components meet in the mixture, their combined inhibition effect is enhanced. The highest inhibition of growth in both sample series generally occurs in standard samples 1 ST and 2 ST as any fly ash addition into the mixture increases the possibility of algae growth on the surface.

4. Conclusions

Green algae represent one group of all the variety of biodeteriogens that participate on building bio corrosion. For the purposes of testing and evaluation the possibility of algae growth on different concrete surfaces, a new methodology was developed and used for testing 10 different concrete samples. The results show that both the type of cement and the type of fly ash incorporated into the concrete mixture influence the algae growth. The theory that fly ashes exhibiting ecotoxicity should suppress the growth when incorporated into concrete mixtures was disproven.

Acknowledgements

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References

[1] Büchli R and Raschle P 2004 *Řasy a houby na fasádách* (Ostrava: MISE, s.r.o.) p 108 ISBN 978-80-254-8786-0
[2] Wasserbauer R 2001 *Biologické znehodnocení staveb* (Praha: ABF) p 257 ISBN 80-86165-30-2
[3] Ledererová J 2009 *Biokorozní vlivy na stavební díla* (Praha: Silikátový svaz) p 273 ISBN 978-80-86821-50-4.
[4] ČSN EN 15458 (672032) Paints and varnishes - Laboratory method for testing the efficacy of film preservatives in coating against algae