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The Influence of Micromycetes on the Corrosion Behaviour of Metals (Steel, Al) under Conditions of the Environment Polluted with Organic Substances

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Biocorrosion is a process in which metabolic activities of microorganisms supply insoluble products that accept electrons from the base metal. The aim of this study was to determine the influence of 5 species of mitosporic fungi (*Chrysosporium merdarium*, *Penicillium cyclopium*, *Arthrinium phaeospermum*, *Cladosporium herbarum*, *Aspergillus niger*), obtained from metals exposed to natural environment conditions for a long time (0.5 to 2 years) on steel and Al, which are the main constituents of construction materials of technical purpose used in the polluted environment. The investigation were carried out in the following directions: fungal interaction with metal (steel, Al) surface; products of interaction of metal with fungi; metal surface morphology after the impact of fungi: nanometric evaluation of changes in the surface of metals. The influence of fungi on the corrosion rate was evaluated using polarization resistance as a criterium.

Keywords: steel, aluminium, micromycetes, corrosion, environmental, pollution, organic substances.

1. INTRODUCTION

Fuel and other liquid substances of technical purpose are kept in reservoirs of various volumes and are pipelined over pipes of different diameters produced from various alloys of steel or aluminum. The pipes soon become polluted with an organic film, where microorganisms start to thrive promoting corrosion processes on the surface of metallic equipment. This hampers the exploitation of the equipment, shortens their useful life, pollutes the stored products and the environment [1].

In the case of metallic materials, undesirable changes in material properties due to a biofilm (or biofouling layer) are referred to as biocorrosion or microbially influenced corrosion (MIC). Biofouling and biocorrosion occur in terrestrial and aquatic habitats varying in nutrient content, temperature, pressure and pH. Interfacial chemistry in such systems reflects a wide variety of physiological activities carried out by diverse microbial populations thriving within biofilms. Biocorrosion can be viewed as a consequence of coupled biological and electron-transfer reactions, i. e. redox reactions of metals, enabled by microbial ecology. Microbially produced extracellular polymeric substances (EPS), which comprise different macromolecules, mediate initial cell adhesion to the material surface and constitute a biofilm matrix. Despite their unquestionable importance in biofilm development, the extent to which EPS contribute to corrosion is not well understood [2, 3].

The study of microbial interactions with metallic materials led recently to the formulation of a unifying electron-transfer hypothesis of biocorrosion, using MIC of ferrous metals as a nodal system [4]. According to this hypothesis, biocorrosion is a process in which metabolic activities of microorganisms supply insoluble products that can accept electrons from the base metal. This sequence of biotic and abiotic reactions produces a kinetically favoured pathway of electron flow from the metal anode to the universal electron acceptor, oxygen.

The role that the organic component, i. e. the biofilm matrix, plays in the electron transfer processes has not been considered in the unified electron transfer hypothesis, despite evidence that enzymes active within the biofilm matrix and metal ions bound by EPS can catalyze cathodic reactions [5]. It is generally acknowledged that microbial EPS are a complex mixture of macromolecules, such as proteins, polysaccharides, lipids and nucleic acids, and that their composition changes with microbial species, physiological status of the cells, and a wide range of environmental factors [6, 7].

Biocorrosion processes at the metal surface are associated with microorganisms, or the products of their metabolic activities, which are characteristic of different types of microorganisms. Fungi can survive as spores in hydrocarbons in the absence of water and germinate when water is available. In addition to water all organisms require carbon, nitrogen, phosphorus, sulfur and other trace elements for growth. Fungi can use many organic and inorganic materials as sources of nutrients and energy. The first step in fungal decomposition of hydrocarbons requires molecular oxygen and the products are alcohols, aldehydes and aliphatic carboxylic acids. Acids produced by fungi are damaging to metals and other materials [8 – 10]. The acids most frequently cited as being produced by fungi are formic, citric and acetic. H_2SO_4 at pH 1.5 exhibits a corrosion current of 3.86 mA cm^{-2}, which is a little more than twice that for formic acid. For the 1 % organic acid solutions, the corrosion currents for the organic acids ranged from 0.27 mA cm^{-2} to 0.03 mA cm^{-2} and the H_2SO_4 at pH 3.0 exhibits a corrosion current of 0.27 mA cm^{-2}. These are generally lower than for concentrated solutions.

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The magnitude of the corrosion currents is sufficient to produce significant corrosion of steel [11, 12]. Some authors [13 – 15] showed, that the inhibition action of fungi to Fe and Al corrosion lies primarily in increase of charge transfer resistance through the inner oxide layer. MIC acceleration was ascertained for zinc while corrosion inhibition was typical of aluminium.

The monitoring of wet and dry deposition rates of pollutants (air born Cl–, sulphur and nitrogen gaseous compounds and aerosols), time of wetness, identification of fungal species on the metal surface and in precipitations collected from the samples were carried out in the different regions (marine, rural, urban) of Lithuania. Several fungal species able to survive or adapt to the metal substrata were detected on all the metals studied [16].

The strain of *Aspergillus niger* Tiegh. was isolated from metal samples. The comparison of the QCM data between the samples affected by *A. niger* and abiotic ones showed a marked increase in the electrode mass due to the development of a biofilm and microbially influenced corrosion. The study demonstrated a unique capability of QCM to sense microbiological corrosion *in-situ* providing continuous and sensitive data on the mass changes during a long-term metal subjection to the influence of microorganisms [17].

Biodeterioration of metals is a fairly new field in microbiology and not accessible to conventional microbiological methods. Suitable techniques for their investigation have been developed only recently and the corrosion relevant biochemical and microbiological parameters first have to be identified, because influence of microorganism on corrosion kinetics depends upon the physico-chemical conditions at the interface, such as the pH value, oxygen concentration, redox potential, water content and ionic strength. As the microorganisms adhere to the corroding surface, by means of their physiological activity they will be able to change all of these parameters in the most corrosion relevant way, i.e. directly at the interface [18].

The aim of this study was to determine the influence of 5 species of mitosporic fungi, obtained from metals exposed to natural environment conditions for a long time (0.5 to 2 years) on steel and aluminium, which are the main constituents of construction materials of technical purpose used in the environment polluted with organics.

2. MATERIALS AND METHODS

Five species of microscopic fungi were selected for studies: 1. *Chrysosporium merdarium* (Link ex Grev.) J.W.Carmich. (Syn. *Sporotrichum merdarium* Link ex. Grev. and many others) are detected in soils on the vegetal remains, technical substances, epoxy glues, aromatic polyamides, polymetafenilenizoftalamides, SiO_2 with bakelite tars, polyamides, fluorine compounds and other substrates. The fungi of this species actively destroy pektine, chitine and proteins. The optimal growth temperature is +20 ºC, the minimal one being +7 ºC and the maximal one approximately +37 ºC. The fungi often dyes the substrate yellow, growing on same substrates abundantly produces citrinin (=antimycin) $C_{13}H_{14}O_5$, which belongs to the benzopyran-3-carboxylic acids group of substances. Citrinin is a mycotoxin of nephrotoxic and carcinogenic effect.

2. *Penicillium cyclopium* Westling (many synonyms are known) micromycetes grow rapidly: after 10 days colonies may be as great as 4.5 cm – 5 cm in diameter, they are common in soils of all the continents on the vegetal remains, trees, timber in the rhizosphere of plants, especially abundantly they are present in the roots zone of halophyte plants. The fungi actively destroy substrates rich in lignine and cellulose, are detected on various polymeric materials. They are able to destroy tanines and actively produce organic acids (ciklopiazonic, penicillic, puberulic, citric, puberulonic), which allow them to develop on metals and affect their corrosion behaviour during exploitation. They develop in substrates of various acidity (pH 2.0 – 10.0), osmophiles, the most rapidly grow at a humidity of 81 % – 84 %. The fungi release into the environment a variety of secondary metabolites, which are often toxic and allergic.

3. *Arthrinium phaeospermum* (Corda) M.B.Ellis Syn. *Papularia sphaerosperma* (Pers. ex Gray) Höhn fungi are widely spread in the world they are detected on vegetal remains, soils, often on other substrates of natural and synthetic origin. The optimal growth temperature is about 20 ºC, minimal – about 8 ºC, maximal about 30 ºC. They produce some specific substances, for example the tetrahydroantrakvinonic pigment – bostucine, which is regarded as a secondary metabolite close to altersolanoles, succine acid, ergosterol, fenole compound and α-threo-β-hydroxiasparte acid distinguished for its antibiotic activity.

4. *Cladosporium herbarum* (Pers.)Link ex. Gray fungi are spread in all the continents on various substrates. They are very resistant to external factors, they can grow under extremal conditions, lacking light, nutrition, under the action of chemical substances and other unfriendly factors. The optimal growth temperature is about +22 ºC, minimal about 0 ºC, maximal – about +35 ºC, conidia remain vital even at temperatures higher than +60 ºC. The fungi adapt to salty environment without difficulty, actively utilize pectine, cellulose, lignine, tars, resins and a variety of other compounds. The fungi of this species are very different both from morphologic and functional points of view. Main metabolitic products senataited by the fungus: proteins, organic acids, polysacarrides. The fungi are used in biotechnology to produce steroid preparations (pregnalon and progesterone) and allergens.

5. *Aspergillus niger* Tiegh. – colonies are formed rapidly, they synthesize and release into environment a great variety of different metabolites, their functional activity is peculiar and allows them to assimilate various substrates and spread in the environment. The fungi actively produce various acids: kojic, citric, oxalic, phenylacetic, indolylacetic, dihydroxydibenzenecarboxylic, glutaconic, 4-hydroxymandelic acid and many others. It should be mentioned that these fungi are distinguished for their ability to detoxicate aflatoxins, which are synthesized by other fungi belonging to this genus. The majority of strains of these fungi are toxic to warm-blooded animals, they often cause mycosis in humans, animals and birds. They actively grow and develop at temperatures of +35 ºC and higher, however they can be found even at a low temperature of +2 ºC.

Plates (100×150×3 mm) of the two principal metals: low carbon steel (LCS) (C 0.05 % – 0.12 %, Cu 0.003 % – 0.10 %, P <0.07 %) and Al (Al >99.5 %) were used in these investigations. The exposure metal plates were cleaned with a fine suspension of $Mg(OH)_2$ and high purity acetone to minimize the initial contamination of the surface by nutritive substances from the environment. To determine the capacity of micromycetes to adapt to metals, the experiments were performed under laboratory conditions using a malt agar extract poor in nutritive materials [14, 18 – 21].

The contact of the metal with fungi was investigated: metal plates were placed in Petri dishes filled with a sterile agar medium of malt extract supplied with chloramphenicol (50 mg l^{-1}). After that the medium with metal was sown up with the fungi of 5 above-mentioned species: 1. *Chrysosporium merdarium* (Ch_1), 2. *Penicillium cyclopium* (P_2), 3. *Arthrinium phaeospermum* ($Arth_3$), 4. *Cladosporium herbarum* (C_4), 5. *Aspergillus niger* ($A.n._5$), 6. The medium with metal plates was not contaminated with fungi (reference – K_2). 7. Metal exposed to common conditions (room temperature and common humidity) was not contaminated with fungi (reference – K1). The medium with metals was cultivated in a thermostat at a temperature of 26 ºC ±2 ºC. The intensity of fungi growth and metal oxidation and surface changes were evaluated after 15 and 30 days. The extent of fungal growth was assessed by the naked eye and light microscopy in accordance with the scheme:

– no fungal growth observed on specimens under the light microscope – 1 point;

– mycelium which branched hyphae and possibly sporulation, visible under the light microscope – 2 points;

– growth of fungi, spare but visible to the naked eye under the light microscope, sporulation clearly visible – 3 points;

– growth of fungi clearly evident but covering <25 % of tested surface – 4 points;

– heavy growth of fungi visible to the naked eye and covering >25 % of the surface – 5 points [21].

A scanning electron microscope EVO 50 EP (Carl Zeis SMTAG, Germany) was used to characterize the morphology of the metal surface.

A Nicolet model 5700 Fourier transformation infrared spectrometer (FTIR) in conjunction with a 10 Spec-(10 Degree Specular Reflectance Accessory) was used for spectroscopical investigations of corrosion products on metal plates. Samples were placed on reflectance accessory and spectra were obtained in the reflectance mode. In all cases the spectral range was 4000 cm^{-1} – 400 cm^{-1} (reciprocal wave length), with a 4 cm^{-1} resolution and 64 scans. Spectral manipulation included baseline correction, removal of carbon dioxide absorption bands and subtraction of water vapour interferences. The samples of corroded metal plates were cleaned with dry filter paper. As a reference sample, the metal plate stored to 30 days without biological intervenes was used. The atmospheric corrosion products formed on metal plates were eliminated, collected and their FTIR spectra were recorded. In this way the products formed by biological agents were detected [22].

The changes in polarization resistance of metals plates were determined from electrochemical impedance spectroscopy (EIS). The EIS measurements were carried out using an electrochemical glass cell equipped with holes for a metal specimen as well as Ag/AgCl reference and Pt counter electrodes. The metal sample was mounted in a special holder, placed in the cell filled with 3.5 % NaCl and EIS measurements were started after 5 – 10 min. These studies were performed at open circuit potential applying a signal of amplitude of $\Delta E = \pm 5$ mV. A P/G/FRA Autolab 302 apparatus (The Netherlands) was used.

One of the most promising techniques for the characterization of microbial adhesion and metal surface interaction is AFM. This method uses atomic force microscopy (AFM) operating in force mode, which offers both imaging capabilities and quantitative measurements of forces between the AFM tip and the sample. The surface morphology of Al and steel (Fe) samples after impact with 5 different fungi was examined with a scanning probe microscope Exlorer (VECO Topometrix, USA). The determined R_q – root-mean Square Roughness (rms) nm. The average roughness is the area between the roughness profile and its mean line or the integral of the absolute value of the roughness profile height over the evolution length. R_q – is a statistic parameter, which shows the average square roughness of the metal surface and it is expressed in nm.

3. RESULTS AND DISCUSSION

3.1. Fungi interaction with metal surface

Fungi play an important biogeochemical role in the biosphere and are intimately involved in the cycling of elements and transformation of both organic and inorganic substrates. They are ubiquitous members of subaerial and subsoil environments, and often become a dominant group in metal-rich or metal-polluted habitats. Fungi have ability for growth under extreme environmental conditions which allows successful colonization of metal surfaces and other metal-rich habitats [23].

After visual evaluation of the interaction of the surface and fungi, it can be stated that the fungi studied contaminated steel plates differently (Fig. 1, Table 1). The mycella of *Chrysosporium merdarium* (Fe_1) fungi condensed on the interface with steel plates, in places the threads of micellium spread over the metal surface (Fe_1). The micelium of *Penicillium cyclopium* fungi changed at the interface, in places grew on the edges of plates, fungi conidia were strongly attached (Fe_2). The micellium of *Arthrinium phaeospermum* fungi gradually covered the surface of steel plate, at the beginning the colour changed, darkened, especially at the substrate (Fe_3). The fungi of *Cladosporium herbarum* species formed colonies straight on the surface of steel plate (Fe_4). The *Aspergillus niger* fungi covered about two thirds of the surface, the film was formed of attached conidia and large extract drops on spreading micellium were seen (Fe_5).

On the surface of aluminium after 30 days of exposure formed colonies of *Cladosporium herbarum* fungi were clearly seen, in places the colonies were formed straight on the metal surface, there were abundant small and large

1. *Chrysosporium merdarium*

2. *Penicillium cyclopium*

3. *Arthrinium phaeospermum*

4. *Cladosporium herbarum*

5. *Aspergillus niger*

K_2 – without Fungi

Fig. 1. A general view of mycromycetes growth on malt extract with steel and Al after 30 days of growth at 26 ±2 ºC (in colour on-line)

extract drops colored yellowish and dark reddish chestnut colour (Al_1). The conidi *Penicillium cyclopium* were spread over the surface of aluminium plate, a clear development of the mycelium was observed only on the edges of metal plates (Al_2). The mycelium of *Arthrinium phaeospermum* fungi covered the whole surface of aluminium plates (Al_3). The colonies of *Cladosporium herbarum* fungi were distinctly formed only in discrete places of the interface of metal with medium (Al_4). *Aspergillus niger* developed on the edges of metal and only in some places of metal surface it was possible to observe the beginning of fungi development (Al_5).

The analysis of data of the changes in steel and aluminium plates visible to the naked eye after 30 days of exposure under laboratory conditions at a temperature of (26±2) ºC has shown (Fig. 2) that the steel surface changed

Table 1. Reaction of steel and Al metals to the impact of micromycetes

Fungal species	Changes on metal surface	Intensity of deterioration in points (1 – 5)
Iron (Fe) after 30 days of exposure to fungi		
Chrysosporium merdarium	The Fungi intensively grew on the edges of a metal plate, less intensively in the centre, the mycelia are yellow	3
Penicillium cyclopium	Fungus covers only the edges of plates, conidia are scattered over the whole plate surface	2
Arthrinium phaeospermum	Fungus covers the edges and the micellium grows like a cobweb towards the centre	2
Cladosporium herbarum	Fungus colonies are formed on the surface of a plate, their development is restricted	4
Aspergillus niger	The plate surface is covered by a layer of scattered conidia and thin micellium and is heavily corroded	4
Plates were exposed to a medium not contaminated with fungi (K_2)	Fungi did not grow on a metal surface	0
Aluminium (Al) after 30 days of exposure to fungi		
Chrysosporium merdarium	The metal is covered by a compact mycelia, denser at the edges, thinner in the center, moisture accumulates intensively	3
Penicillium cyclopium	Adhered fungus conidia are seen on the plate surface, micellium covers the surfaces in places	2
Arthrinium phaeospermum	The fungus micellium covered the whole plate surface	5
Cladosporium herbarum	Fungus colonies are formed in certain places on the edges	2
Aspergillus niger	The plate edges are covered with a thin layer of fungi micellium	2
Plates were exposed to a medium not contaminated with fungi (K_2)	Fungi did not grow on a metal surface	0

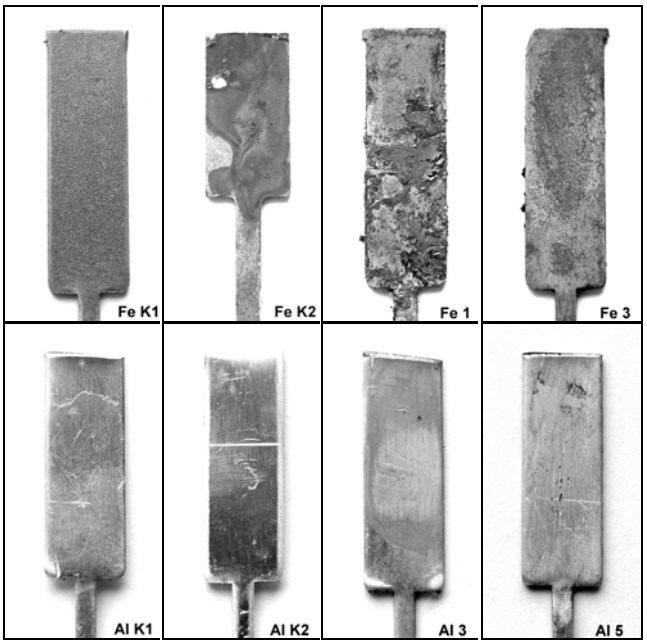


Fig. 2. More pronounced changes in steel and Al surfaces after 30 days of contact with developing fungi of various species: K_1 – not exposed plates; K_2 – exposed without fungi; Ch_1 – contaminated with *Chrysosporium merdarium*; P_2 – contaminated with *Penicillium cyclopium*; $Arthr_3$ – contaminated with *Arthrinium phaeospermum*; Cl_4 – contaminated with *Cladosporium herbarum*; $A.n._5$ – contaminated with *Aspergillus niger* after 30 days of exposure at 26 ºC ±2 ºC

most under the action of *Chrysosporium merdarium* (Fe_1) and *Arthrinium phaeospermum* (Fe_3) fungi, while the surface of aluminium changed most under the action of A. *phaeospermum* (Al_3) and *Aspergillus niger* (Al_5). The single threads of micellium of the former fungus were tightly attached to the Al plate surface, the Al surface under action of the latter fungus became rough.

3.2. Products of metal surface and fungi interaction

The nature of the organic substances remaining on the surface of steel and Al specimens was determined by using the surface reflection spectroscopy method. The FTIR spectra of the studied samples exposed for 30 days to the above described conditions are presented in Figs. 3 – 7. The FTIR spectra of remaining substances of the reference (K_2) on Fe and Al plates after 30 days of contact were close to those of the plates contaminated with *Chrysosporium merdarium* fungus. In corresponding products on the steel surface amide groups dominated, while on the Al surface a S–OH group was detected. At that time C–C deformation fluctuations in both cases were virtually identical (Fig. 3).

After 30 days of the contact of steel samples with *Penicillium cyclopium* fungi the attempts to detect an organic substance on the surface of the plate from the FTIR spectrum failed, while on the surface of Al specimen the traces of humidity –OH groups and various organic compounds (organic acids, indole alkaloids, trichothecenes et al.) were detected.

The spectra of organic substances after the analysis of the effect of *Arthrinium phaeospermum* on Fe and Al were similar: an intensive peak of –OH group and a common set of the remains of organic substances $–CH_3$ bands, –C–C–, –CH groups. After the action of *Cladosporium herbarum* on Fe the traces of organic substances were recorded and absorption bands on the Al surface were observed. The spectra were expressed by weak peaks, at the same time after the action of *Aspergillus niger* an intensive peak

–C–H on iron was observed, as well as a great set of deformed vibrations in the area of 600 nm^{-1} – 400 nm^{-1} on Al (Fig. 7). It should be noted, that scanty spectra of organic substances on Fe plates after the influence of *Penicillium cyclopium* and *Cladosporium herbarum* can be a sequence of the fact that the products of the activity of these fungi are veiled by the layers of Fe oxides and hydroxides, which emerge on the surface of the basis and the sound does not reach the purpose, as it strays in catastrophically rough irregularities of the surface of Fe basis.

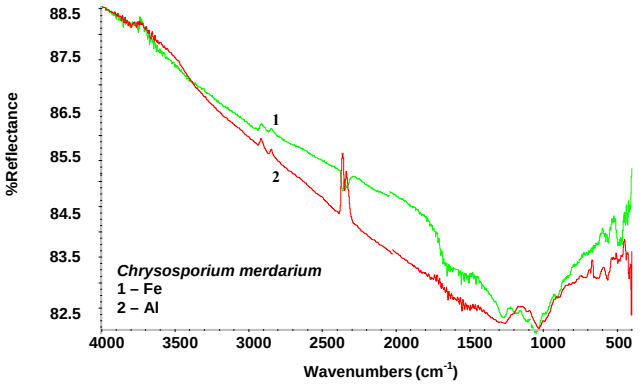


Fig. 3. FTIR spectra of steel and Al specimens exposed to *Chrysosporium merdarium* fungi for 30 days

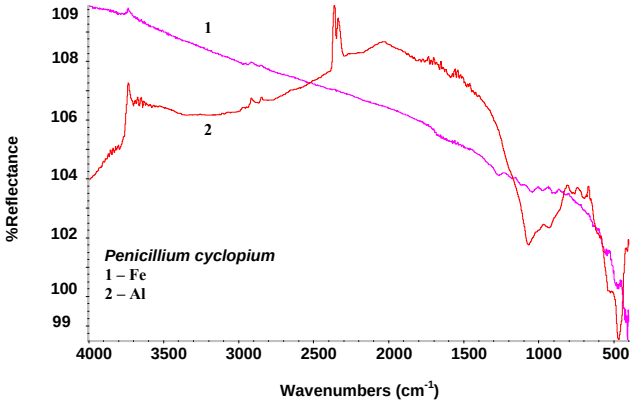


Fig. 4. FTIR spectra of steel and Al specimens exposed to *Penicillium cyclopium* fungi for 30 days

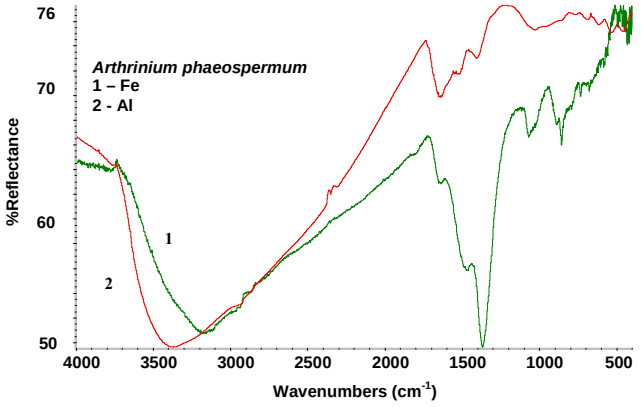


Fig. 5. FTIR spectra of steel and Al specimens exposed to *Arthrinium phaeospermum* fungi for 30 days

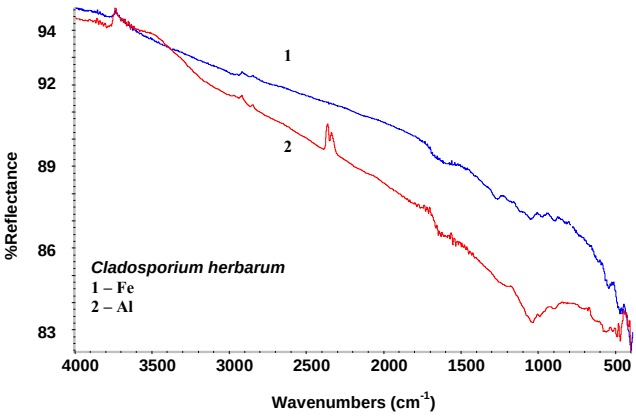


Fig. 6. FTIR spectra of steel and Al specimens exposed to *Cladosporium herbarum* fungi for 30 days

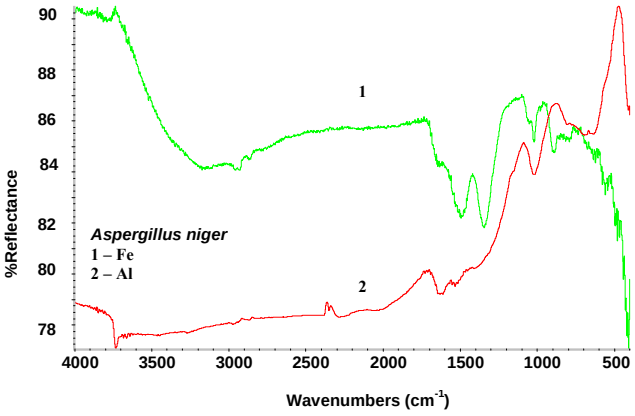


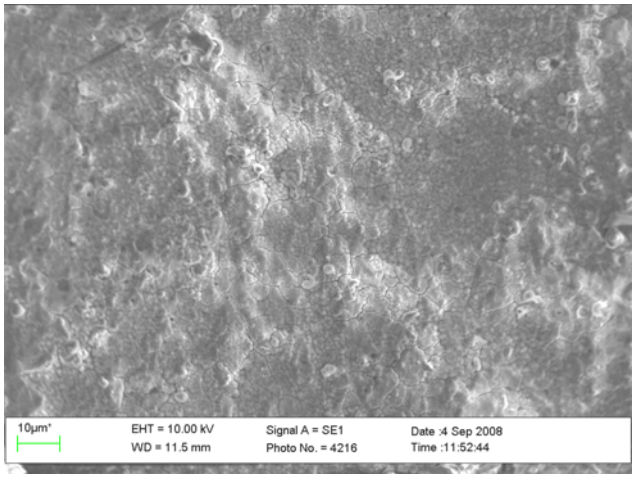
Fig. 7. FTIR spectra of steel and Al specimens exposed to *Aspergillus niger* fungi for 30 days

3.3. Metal surface morphology after fungi impact

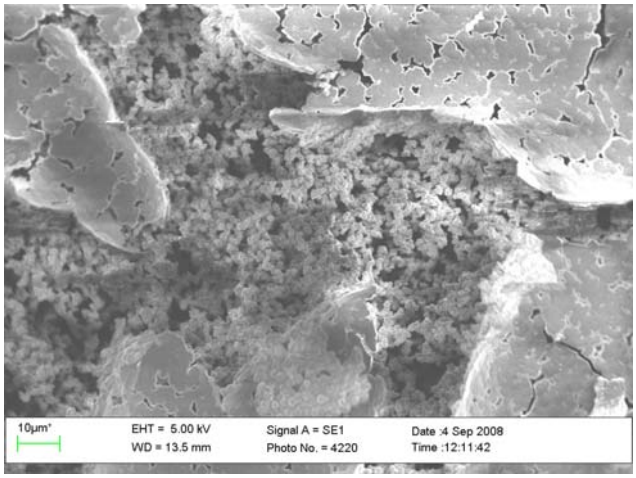
The morphological changes in steel and aluminium basis were studied by the SEM and AFM methods. The SEM method made it possible to analyze surface changes related to the biocorrosive damages in the micrometer range. Images were analyzed by implementing of isograms "Image J". In this basis the linear values and Fereti diameter of the whole picture were determined. It should be mentioned, that the irregularities of surface relief was a handicap to calculation of Fereti diameter of steel samples. The photos of Fe and Al plates surfaces without contact with fungi ($K_1 - K_2$) and after 30 days of contact of metal plates with different fungi (1 – 5) are given below.

As seen from the data in Fig. 8 the changes in the basis of steel plates surface are different. The surface of the specimen exposed to the medium without fungi (Fe (K_2)) is covered with single spots of corrosion, whose diameter is 2 μm – 3 μm. Whereas the surface of steel after its contact with *Chrysosporium merdarium* (Ch_1) is heterogeneous, formed wind creaks continuous triangled hinges (2 μm – 4 μm) and film cracks, most probably of iron oxide, finely dispersed corrosion products (1 μm – 2 μm) are observed (Fe(Ch_1)).

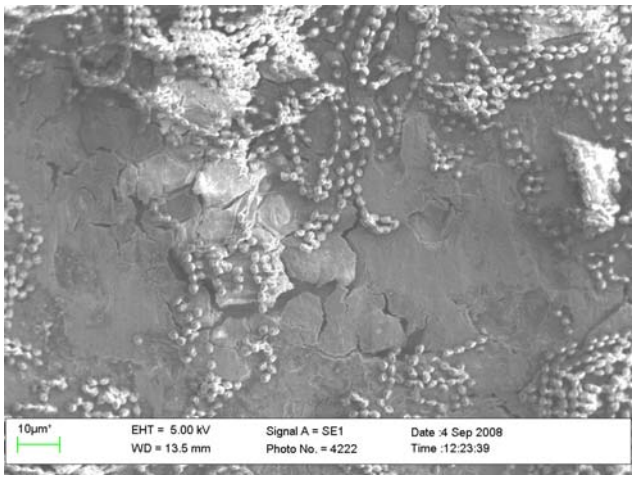
On the steel surface exposed to the medium with *Penicillium cyclopium,* 10 μm – 15 μm Fe oxide scales with deep slits between them are seen. Attached fungi



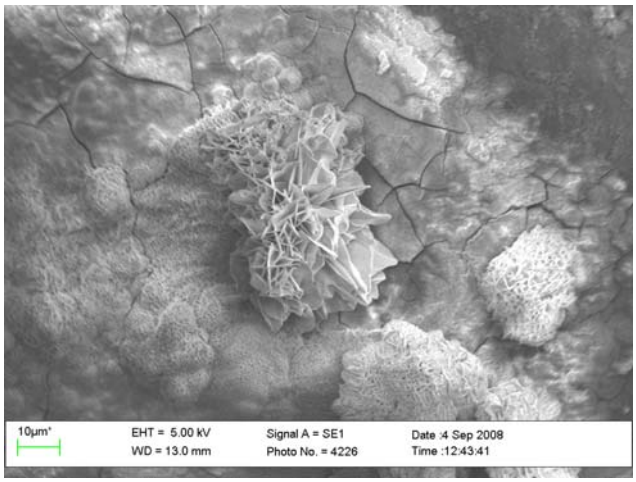
Fe K_2



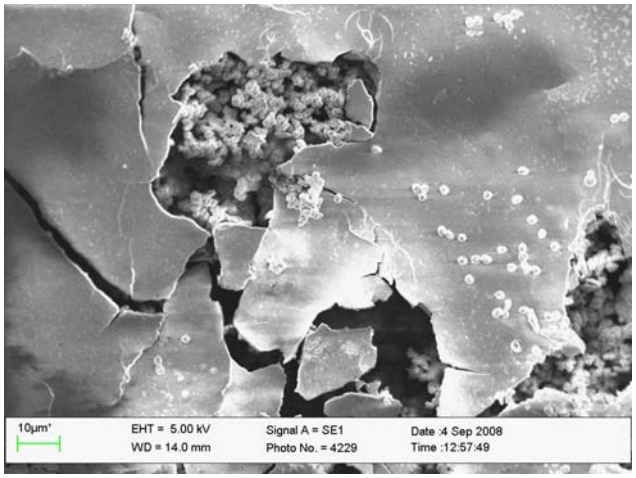
Fe_1



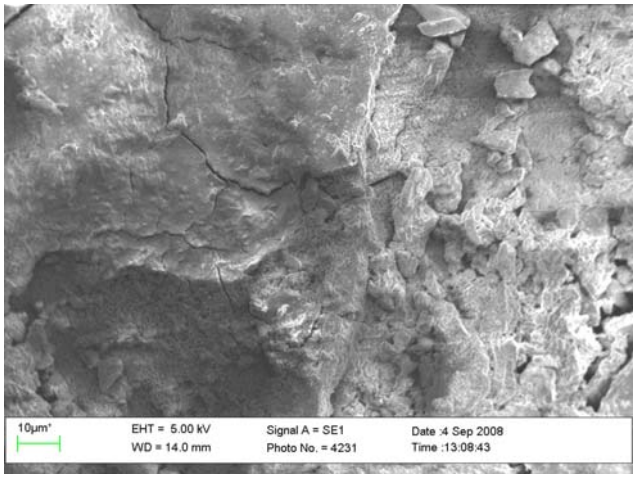
Fe_2



Fe_3



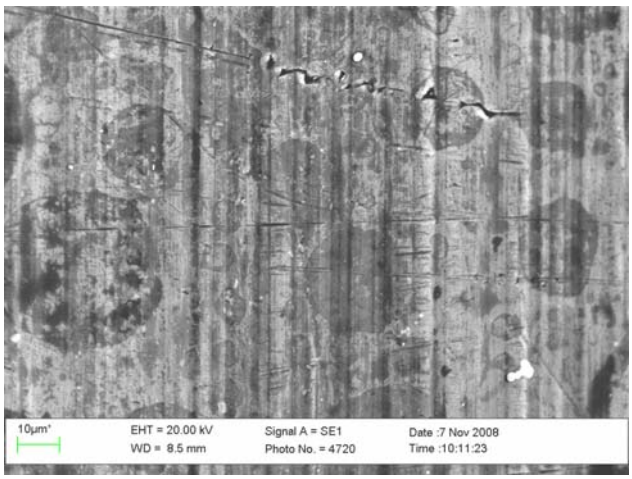
Fe_4



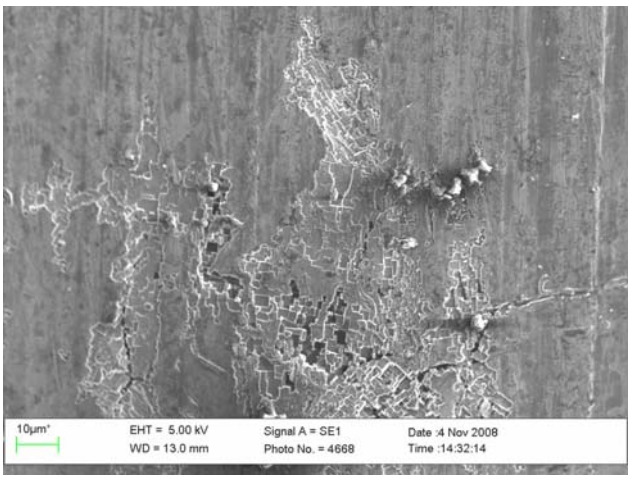
Fe_5

Fig. 8. A general view of steel surface after 30 days of exposure to a nutrient medium at 26 ºC ±2 ºC: K_2 – without fungi; Fe_1 – contaminated with *Chrysosporium merdarium*; Fe_2 – contaminated with *Penicillium cyclopium*; Fe_3 – contaminated with *Arthrinium phaeospermum*; Fe_4 – contaminated with *Cladosporium herbarum*; Fe_5 – contaminated with *Aspergillus niger*

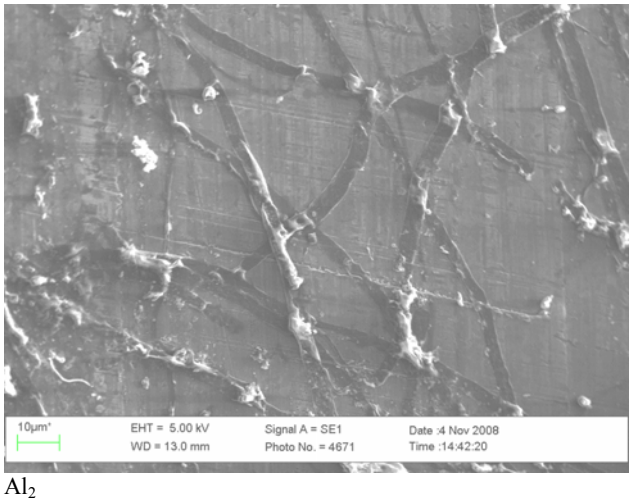
conidia chains of the same size (~2 µm) are seen on the scales, in places forming small colonies. Under the influence of *Arthrinium phaeospermum* the surface of the basis Fe plate was covered with iron oxide scales of 15 µm – 20 µm in places drifts zones of scales of 5 µm – 7 µm are observed, crystalline formations of toothed form, whose needles reach 2 µm – 3 µm in length and up to 0.5 µm in thickness, as well as 5 µm – 10 µm plates arranged in a fan-shaped manner. They are possible Fe crystal hydrates formed when corrosion products were dried out after the influence of fungi on the surface of the plate. Rather thick ~0.5 µm Fe oxide layers whose



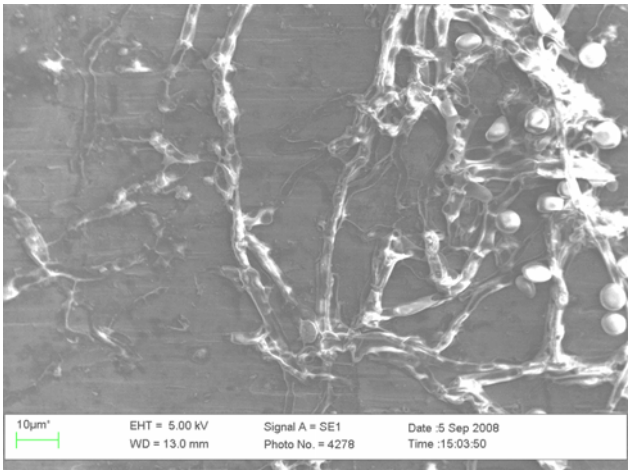
Al K_2



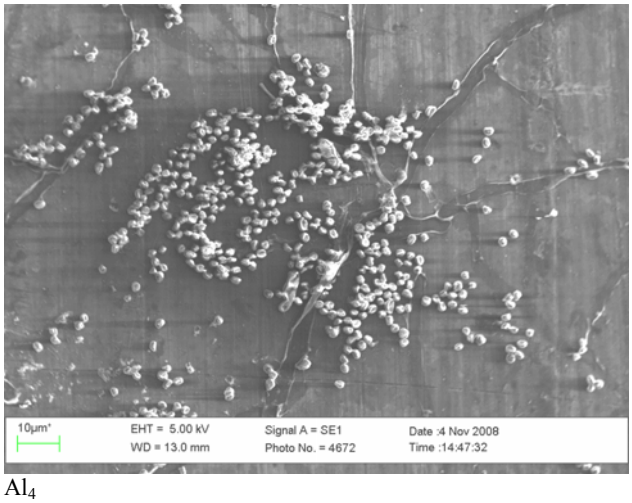
Al_1



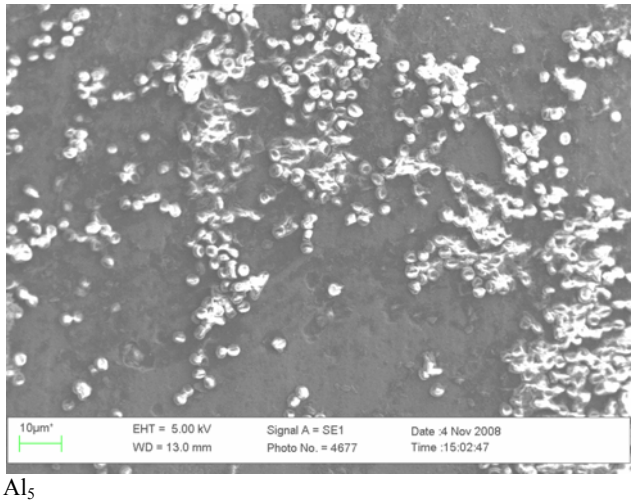
Al_2



Al_3



Al_4



Al_5

Fig. 9. A general view of aliuminium surface after 30 days of exposure to a nutrient medium at 26 ºC ±2 ºC: K_2 – without fungi; Al_1 – contaminated with *Chrysosporium merdarium*; Al_2 – contaminated with *Penicillium cyclopium*; Al_3 – contaminated with *Arthrinium phaeospermum*; Al_4 – contaminated with *Cladosporium herbarum*; Al_5 – contaminated with *Aspergillus niger*

structure is brokevbn and which contain fine-grained 0.5 µm – 2.0 µm conglomerates of corrosion products are observed among scales. Conidia of 2 – 3 µm in size, whose morphology distinctly differs from that of iron oxide derivatives seen in splits, are observed on the oxide surface in the right. Under the influence of *Aspergillus niger* the surface of the plate basis becomes porous and chinky, there are a lot of decomposition remains, along with the scales and crumbling observed on the surface, finely spongy structures not greater than 0.5 µm are seen. The pores are located chaotically on the whole surface of spurs.

The changes in the surface of aliuminium plates under the influence of microsporic fungi were different from those in steel plates (Fig. 9).

On exposed Al plates without fungi on the surface (AlK_2), massive corrosion seats of 5 μm to 30 μm in diameter are observed. Most probably it is a formed layer of aluminium sulphide and oxide.

On the surface of aluminium plate, affected by *Chrysosporium merdarium* fungi a net of 2 μm – 3 μm threads with knots and spatial structure up to 5 μm – 7 μm away from the Al surface are observed. This is a micellium of attached fungus and its derivatives (Fig. 9, Al_1).

The view of the surface of aluminium plate affected by *Penicillium cyclopium* is similar (Fig. 9, Al_2) to that of steel plate surface affected by the same fungi (Fig. 8, Fe_2), just the thready structure here is flatter, more adherent to the Al basis and with corrosion traces (Fig. 9, Al_2). In the left-hand bottom corner of the figure accumulation of irregularly-shaped destroyed remains of biological origin is observed.

On the surface of specimen exposed to the medium with *Arthrinium phaeospermum* 2 μm – 4 μm thready structures of unique deep bowl appearance in which derivatives of oval to ellipse shape and 3 μm – 5 μm in size similar to fungi conidia are observed (Fig. 9, Al_3). On the surface of Al little ulcers of 1 μm – 3 μm of irregular shape are seen. It can be stated that the thready structure observed on the surface is the result of the action of fungi.

The micellium of *Cladosporium herbarum* herbarium fungi is tightly attached to the surface of Al plate, 2 μm – 4 μm wide corrosion bands are observed as well as fungi conidias surrounding them, which are of oval shape and 1 μm – 2 μm in size (Fig. 9, Al_4). *Aspergillus niger* fungi on the Al plate did not develop deeply and the thready structures did not change, but on the surface there are a lot of adhesed conidias 2 μm – 4 μm in size, under which cellularity and porosity of various shape are observed (Fig. 9, Al_5).

3.4. Nanometric evaluation of changes in the surface of metals (Fe, Al)

Steel surfaces in nanometric level after 30 days of contact with fungi *Chrysosporium merdarium* and *Arthrinium phaeospermum* are presented in Fig. 10. The surfaces of iron plates affected by other fungi were decomposed to such an extent that it was impossible to scan them by the AFM method. The surfaces of reference variants (K_1 and K_2) of iron plates differed significantly. This can be explained by the fact that in both cases there were abundant iron oxide scales on the scanned surface, where humps of various size (200 μm – 300 μm) were usually seen. Under he influence of *Arthrinium phaeospermum* fungi the roughness of the surface of iron plate increased six-fold as compared to that of the reference ($Arth_3$), while under the influence of *Chrysosporium merdarium* (Ch_1) – three-fold.

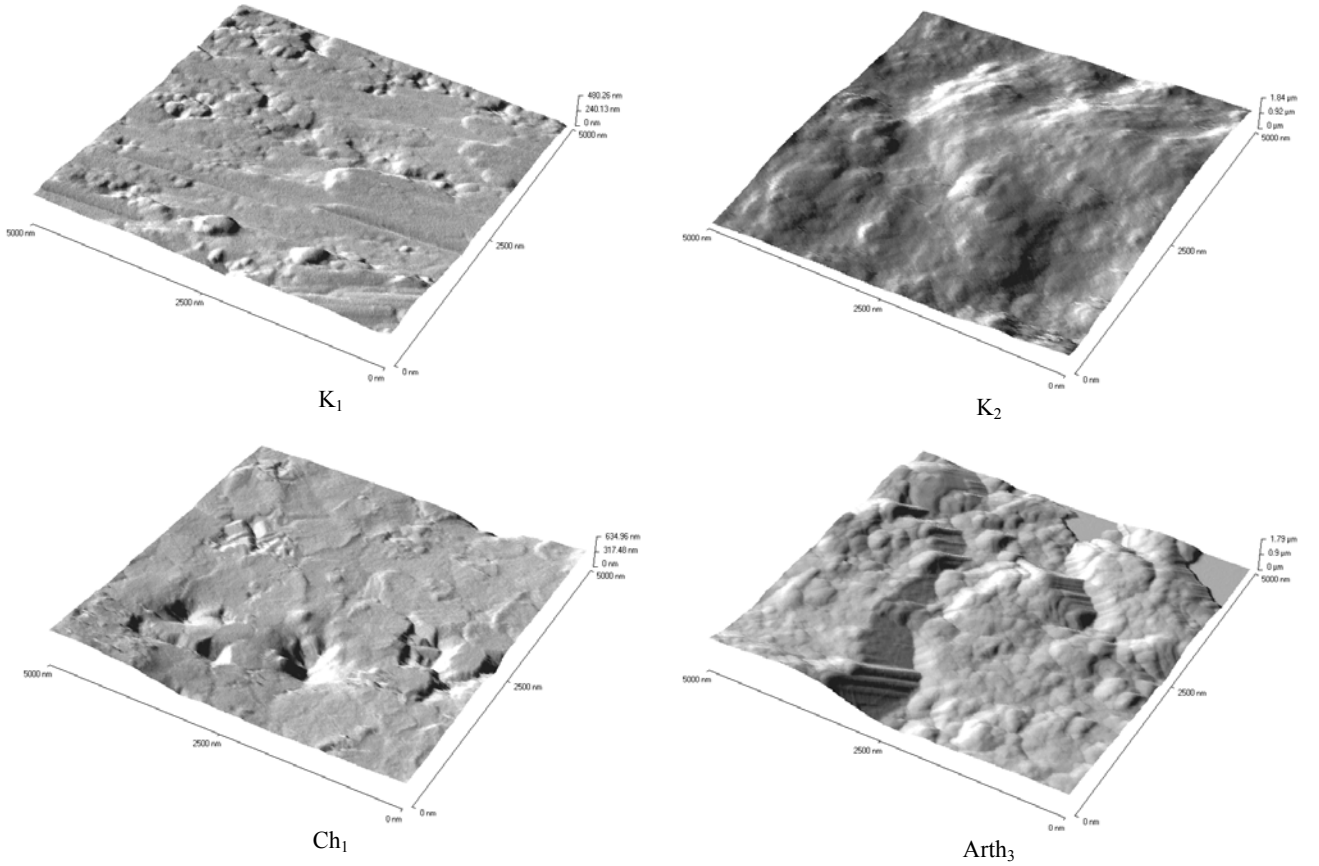


Fig. 10. A morphological view of steel surface after 30 days of contact with developing fungi of various species: K_1 – not exposed plates without fungi; K_2 – exposed under the same conditions without fungi; Ch_1 – contaminated with *Chrysosporium merdarium*; $Arth_3$ – contaminated with *Arthrinium phaeospermum*

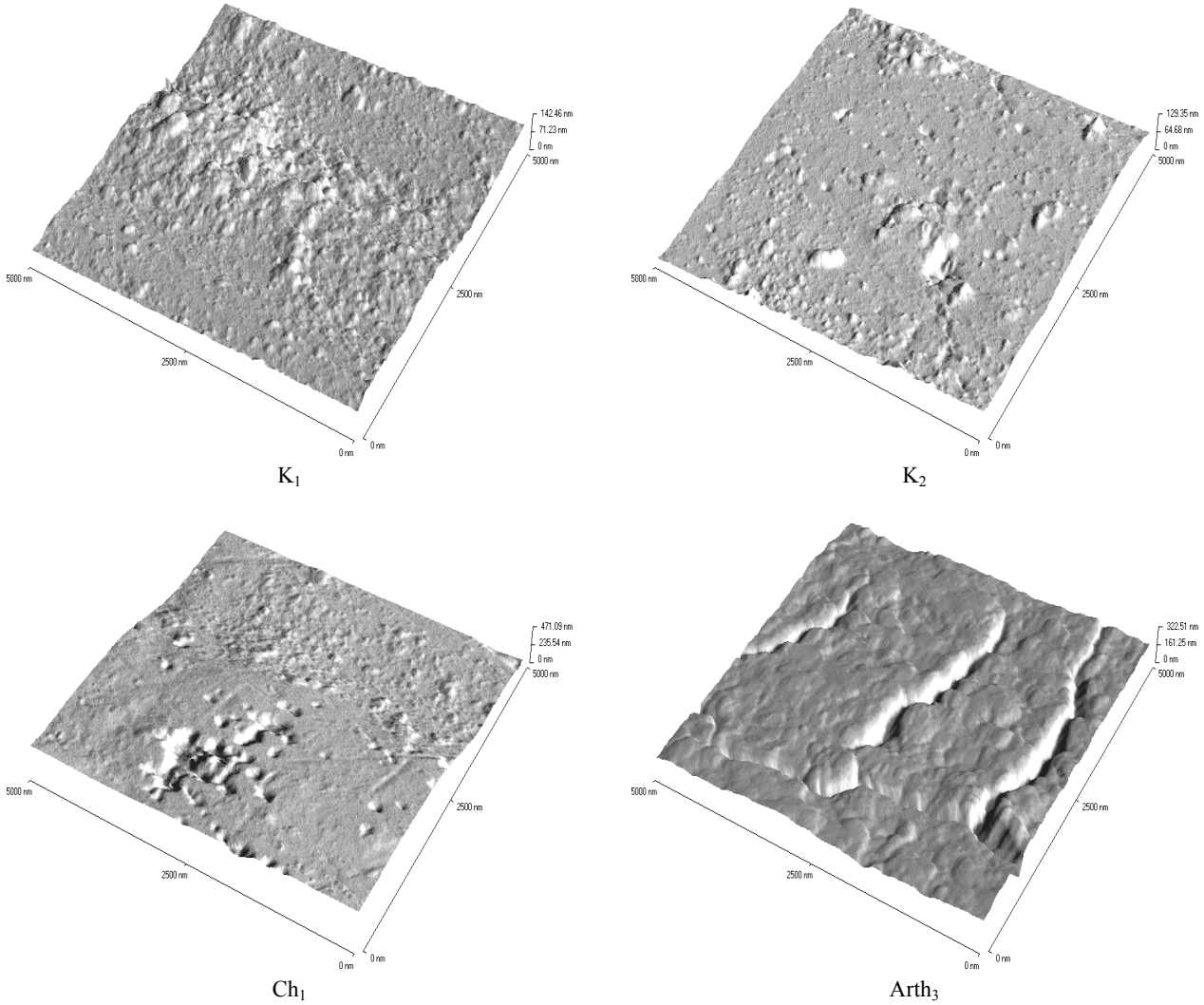


Fig. 11. A morphological view of aliuminium surface after 30 days of contact with developing fungi of various species: K_1 – not exposed plates without fungi; K_2 – exposed under the same conditions without fungi; Ch_1 – contaminated with *Chrysosporium merdarium*; $Arth_3$ – contaminated with *Arthrinium phaeospermum*

Aliuminium surfaces in the nanometric level after 30 days of contact with *Chrysosporium merdarium* and *Arthrinium phaeospermum* fungi are presented in Fig. 11.

These studies are of dotted character the data obtained can be named only by systematic changes in nanomorphology of the steel and Al surface under the influence of the biocorrosion process.

The changes in the average square roughness (rms, R_q, nm) of aluminium surface after 30 days of contact with different species of micromycetes at a temperature of 20 ºC ±2 ºC are presented in Table 2.

From the data presented in Table 2 it is seen that *Chrysosporium merdarium* and *Aspergillus niger* fungi had the most significant influence on the roughness of the basis of Al plate. Partially this was also confirmed by the roughness data measured by the AFM method (Fig. 11). The destruction views of the specimen contaminated with *Arthrinium phaeospermum* fungi particularly be distinguished (Fig. 11, $Arth_3$). On the plate surface longitudinal splits up to 5 μm long and up to 1 μm deep are observed. The deteriorations of a such kind were not detected in other variants of experiment.

Table 2. Changes in the average square roughness (rms, R_q, nm) after 30 days of contact with micromycetes of various species at 20 ºC ±2 ºC

Variant	Steel rms, R_q, nm	Al rms, R_q, nm
K_1 – plates free from fungi kept under dry room conditions	38.45	21.89
K_2 – plates free from fungi kept in contact with a malt agar medium	262.01	32.57
Plates kept in contact with a medium contaminated with 5 various micromycetes:		
1. *Chrysosporium merdarium*	57.45	50.49
2. *Penicillium cyclopium*	*	30.95
3. *Arthrinium phaeospermum*	160.66	35.18
4. *Cladosporium herbarum*	*	31.39
5. *Aspergillus niger*	*	85.44

* – was not measured owing to intensive corrosion.

3.5. Corrosion studies

The influence of fungi on corrosion rate was evaluated using as a criterion the polarization resistance (R_p), which is in inverse proportion to the corrosion current density (j_{corr}). The changes in steel and aluminium plates after 30 days of contact with fungi of various species could be inferred by the data presented in Table 3.

Table 3. Polarization resistance values of steel and Al electrodes in 3.5 % NaCl solution, after impact of different fungi during 30 days of exposure at 26 ºC ±2 ºC

Variant	Polarization resistance R_p	
	Steel, Ω/cm^2	Al, $k\Omega/cm^2$
Reference (K_1)	742.2	122.9
Not contaminated (K_2)	143.8	157.8
Chrysosporium merdarium (Ch_1)	147.2	51.1
Penicillium cyclopium (P_2)	184.6	85.7
Arthrinium phaeospermum ($Arth_3$)	136.5	200.5
Cladosporium herbarum (Cl_4)	145.0	1422.0
Aspergillus niger ($A.n._5$)	143.3	9.0

Polarization resistance changes were determined from EIS data after 30 days of exposure of iron samples to developing fungi of various species. In all cases of steel studies a marked decrease in polarization resistance from its initial value 742.2 $\Omega\,cm^2$, which was determined for a specimen free from fungi, to ~143.3 $\Omega\,cm^2$, which was determined for a specimen contaminated with *Aspergillus niger* fungi. The comparison of the R_p values obtained shows, that the metal surface rather actively corroded, but the formed corrosion products did not possess protective (passivation) properties or they were not clearly pronounced. When steel specimens were exposed to the medium with fungi, very close R_p values, shifting in the interval ~137 $\Omega\,cm^2$ to 185 $\Omega\,cm^2$, which testifies, that fungi did not have any significant influence on steel, or the aggressiveness of the medium was rather high to evaluate the MIC effect.

The polarization resistance of aluminium specimens after contact with growing fungi changed rather differently. Under the influence of the medium free from fungi Al R_p slightly increased from its initial value of ~123 $k\Omega\,cm^2$ up to 158 $k\Omega\,cm^2$, whereas the influence of fungi varied. R_p decreased to 51 $k\Omega\,cm^2$ and 86 $k\Omega\,cm^2$ under the influence of *Chrysosporium merdarrium* and *Penicillium cyclopium* fungi, respectively, whereas the influence of *Aspergillus niger* fungi was more pronounced, because Al R_p only reached 9 $k\Omega\,cm^2$. The results obtained show, that the mentioned fungi can accelerate metal corrosion processes. Especially this was characteristic of *Aspergillus niger* fungi under conditions of this study. However, the influence of *Cladosporium herbarum* was opposite. The mentioned fungi under the same conditions markedly slowed down the rate of Al corrosion. Under the influence of these fungi Al R_p increased up to 1422 $k\Omega\,cm^2$. The R_p value of Al specimen affected by *Arthrinium phaeospermum* increased insignificantly up to ~200 $k\Omega\,cm^2$.

The results of the studies performed show that fungi studied did not have any pronounced effect on the corrosion behaviour of steel. *Cladosporium herbarum* fungi markedly inhibited Al corrosion processes, and the used *Aspergillus niger* strain was distinguished for its strong acceleration properties. This sets one paying attention to different influence of various strain of the same fungi species on metals and their dissimilar reaction to medium factors

Some authors [13, 14] showed, that the inhibion action of fungi on steel and Al corrosion lies primarily in the increase of charge transfer resistance through the inner oxide layer. MIC acceleration was ascertained for zinc while corrosion inhibition was typical of aluminium.

CONCLUSIONS

1. Under the conditions of atmospheric environment polluted with organics, mitosporic fungi is a strong ecologic factor capable of changing polarization resistance, inducing metal oxidation processes, promoting or in some cases stabilizing or decreasing corrosion processes.
2. The influence of various species of mitosporic fungi on metals differs. This depends on the physiological properties, which they inherited, developed and strengthened in themselves under favourable in some cases very specific environmental conditions formed being in contact with certain metals.
3. Under the studied conditions *Arthrinium phaeospermum*, *Aspergillus niger* and *Chrysosporium merdarium* fungi affected steel and Al surfaces most actively. It should be noted that this influence depended on the peculiarities of the metal surface, the level of contamination and the ability of the fungus to develop under extreme conditions formed on the metal surface.
4. There is no point in thinking that metal atmospheric corrosion problems can be studied, assessed and solved positively without biological factors, which often determine the corrosion behaviour of complicated and specific processes, because the subjects of investigation are live organisms which are very sensitive, rapidly react to the changes in the medium, alternate the character of metabolism-synthesize substances inhibiting or promoting corrosion.
5. It should be noted that various strains of the same fungi species have different influence on metals and their reaction to environmental factor differs as well.

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