

Proceedings of the Estonian Academy of Sciences, 2022, **71**, 4, 361–368 https://doi.org/10.3176/proc.2022.4.06 Available online at www.eap.ee/proceedings

ANTIMICROBIAL **MATERIALS**

Microbiological and chemical properties of shungite water

Silver Türk^a, Tarmo Tamm^b, Hugo Mändar^c, Ain Raal^{d*}, Peeter Laurson^e and Uno Mäeorg^e

^a Institute of Biomedicine and Translational Medicine, Faculty of Medicine, University of Tartu, Tartu, Estonia

^b Institute of Technology, Faculty of Science and Technology, University of Tartu, Tartu, Estonia

^c Institute of Physics, Faculty of Science and Technology, University of Tartu, Tartu, Estonia

^d Institute of Pharmacy, Faculty of Medicine, University of Tartu, Tartu, Estonia

e Institute of Chemistry, Faculty of Science and Technology, University of Tartu, Tartu, Estonia

Received 27 March 2022, accepted 9 June 2022, available online 18 November 2022

© 2022 Authors. This is an Open Access article distributed under the terms and conditions of the Creative Commons Attribution 4.0 International License CC BY 4.0 (http://creativecommons.org/licenses/by/4.0).

Abstract. Shungite is used in water filters that remove *Escherichia coli* from water. The mechanism and spectrum of the antibacterial activity of shungite are not precisely known. In this study, shungite and its dried water extract were characterized by means of X-ray diffraction, X-ray fluorescence and iodometry. The dried residue of the water extract of shungite was relatively poor in carbon (28.1% in the rock vs 0.5% in the residue), silica (23.9% in the rock vs 0.3% in the residue) and potassium (1.14% vs 0.05%), but rich in sulfur (1.6% vs 21.6%) and some metals, including iron (1.4% vs 10%), aluminum (2.1% vs 5%) and nickel (0.02% vs 1.14%). The survival of *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Candida albicans*, *Streptococcus uberis* and *Saccharomyces cerevisiae* in shungite water extract, while *Staphylococcus aureus*, *Candida albicans* and *Saccharomyces cerevisiae* survived as well as in distilled water. Neutralization of pH did not abolish the bactericidal effect. However, in the presence of nutrients, shungite water did not show bacteriostatic or bactericidal effects.

Keywords: geomicrobiology, shungite, water, susceptibility, XRD, XRF, beer.

INTRODUCTION

According to legend, keeping shungite rock in water purifies water. Using shungite rocks (Fig. 1) with the intent of purifying water is contemporary practice. Rocks with different properties and compositions may fit under the relatively loose term "shungite".

Shungite is named after the town of Shunga (Karelia) and is an intermediate product between amorphous carbon and crystalline graphite, which contains carbon (30 wt%), quartz (45 wt%), silicate micas (~20 wt%), and various admixtures (V, Ni, Cu, Fe, and oxides thereof; Lyn'kov et al. 2009). In most cases, shungite rocks of different types are distinguishable with the naked eye (Buseck 2002).

Clearly, different types of shungite have different chemical compositions, with the principal shiny and lighter variety having a lower carbon to silica ratio. There has been an interesting debate around the issue whether shungite is a natural source of fullerenes. The topic of fullerenes found in shungite has been addressed elsewhere (Buseck 2002), but it is possible that these results are still only method artifacts (Santos et al. 2016). Moreover, since fullerenes are not water-soluble - water solubility is crucial to our aims of measuring the properties of shungite water - the characteristics of other constituents should be considered more relevant for the present study. The composition of shungite has been reported as 57% SiO₂, 30% C, 4% Al₂O₃, 1.7% H₂O, 1.5% Fe₂O₃, 1.5% K₂O, 1.2% S, 0.6% FeO (Mosin and Ignatov 2013). As concerns the form of carbon, it has recently been suggested to be

^{*} Corresponding author, ain raal@ut.ee



Fig. 1. Shungite. Photo by A. Raal.

fullerene-like or oxygraphene wafers (Sheka and Rozhkova 2014). Particles of 20 μ m have been reported to exhibit the most powerful antioxidant abilities (Skrypnik et al. 2021).

Shungite water extracts and natural shungite water have been researched previously (Charykova et al. 2006). In the water of a river flowing along shungite bedrocks, the ion levels were found not to exceed the criteria for potable water: 0.430 mg/mL Fe, 0.036 mg/mL Al, 0.004 mg/mL Zn, 20 mg/mL Mg, 0.001 mg/mL Ni, 1.9 mg/mL Si, 50 mg/mL Ca, <0.001 mg/mL Cu, <0.0001 mg/mL Cd, and < 0.001 mg/mL Co. On the other hand, shungite sludge water near industrial facilities would definitely exceed the potable water criteria for major ions (2600 mg/ mL⁻¹ Fe) as well as for toxic trace elements (1.9 mg/mL Cd, 4.1 mg/mL Ni). The same study found that shungite infusion and shungite "tea" (short-term incubation with boiling water) have different levels for leaching iron and cadmium into the water phase - shungite infusion contains more cadmium, whereas shungite "tea" contains more iron.

It has been shown in the literature (Buseck 2002; Santos et al. 2016) that fullerenes are not present in dull shungite. On the other hand, when shungites have been studied by high-resolution transmission electron microscopy (HRTEM) (Kovalevski and Moshnikov 2016), signs of fullerene-like structures have been found. According to another source (Sheka et al. 2014), shungite does not contain fullerenes but it contains graphene oxide sheet aggregates that may be more water-soluble due to the presence of hydrophilic groups (oxygen). Perhaps these graphene oxide complexes that share some similarities with fullerenes (large, aromatic and conjugated), as well as some differences (partial oxidation), have an effect similar to that of fullerenes (Sheka et al. 2014). Hence, the presence and role of fullerenes or oxygraphenes in shungite water requires further elucidation.

The biological properties of shungite water have been minimally researched. Shungite has not been studied in vivo in the context of longevity. Fullerenes in olive oil have been reported to double the lifespan of rats (Baati et al. 2012), but such effects were not observed in a recent mouse study (Grohn et al. 2021). It has been previously shown (Mosin and Ignatov 2013) that a shungite filter can adsorb E. coli contamination from water. Incubating 25 mL of water with more than 10⁶ CFU/mL on 15 g of shungite for three days made the water microbiologically clean (Charykova et al. 2006). This has been put into practice: relevant water filters are available in the European Union and Russia. In addition, bacteria adsorbed on shungite have been investigated in the context of oil bioremediation and adsorption of bacteria on shungite particles (Efremova 2006). Animal studies include a dark brown mink study, wherein the injection of shungite water caused no apparent toxicity while displaying antioxidant properties (Tyutyunnik et al. 2009), and a mouse study, wherein the application of shungite reduced inflammation and oxidative damage due to ultraviolet B radiation (Sajo et al. 2017). It has been observed that shungite nanoparticles may adsorb some proteins and alter the redox potential of the environment (Rozhkov and Goryunov 2013). As concerns oxygraphene, a possible constituent of shungite water, 2 hours of incubation with 5 µg/mL, 40 µg/mL and 80 µg/mL of oxygraphene killed 10.5%, 69.3% and 91.6% of E. coli, respectively, with initial concentrations between 10⁶ and 10⁷ CFU/mL (Liu et al. 2011).

Salt content is known to influence the survivability of microorganisms. Already before World War II, it was known that saline water was more antibacterial than distilled water (Ballantyne 1930). A modern mineral water study showed that water with a low mineralization level was actually more antibacterial (Serrano et al. 2012). By the time of the previous study it was already understood that the survivability of microorganisms is affected by temperature, the amount of inoculum, the age of the culture used in making the suspension, whether the bacteria are washed before storing in water, and the presence of additional chemicals in the water. Dallakyan et al. (2018) studied the combined effect of shungite with potassium dichromate and cadmium sulfate on the growth of the culture of green microalgae Scenedesmus quadricauda (Chlorococcales) and concluded that shungite neutralized the toxic effect of heavy metals.

Shungite is used in water filters for water purification. Instead of adsorption, we aimed to measure the antimicrobial properties of shungite water. The other aims of this study were to characterize the chemical composition of shungite water extract, shungite itself and the antimicrobial mechanisms of shungite water, as well as the effect of shungite on fermentation in beer brewing with shungite.

MATERIALS AND METHODS

Preparation of shungite extracts

Particles of shungite rock were obtained from a water purifier column (OÜ Gyllenhaal, Estonia). The water extract for microbiological analyses was prepared by using three parts of shungite rock (150 g) and four parts of distilled water (200 g), heated to boiling and allowed to cool, then infused for 24 ± 2 hours, centrifuged at 1000 g for 5 minutes and decanted. The supernatant (shungite water) was used for determining the antimicrobial properties. The dry mass of shungite water extract made from 150 g of shungite gravel and 200 g of water was 1.2017 g. In addition, cold water extract was prepared from 100 g of shungite gravel and 900 g of deionized water by overnight incubation and subsequent decantation, heating to boiling and cooling to room temperature.

For the preparation of pH-neutralized shungite water, the previously described shungite water was neutralized from pH 2.56 to pH 6.78, controlled by a pH meter (C6010, Consort, Belgium) and a magnetic stirrer. During the neutralization of the shungite water, which itself was clear, the solution turned yellow and gradually a pale brown sediment started to form. After subjecting 50 mL of the yellowish solution to centrifugation in a centrifugation tube, a voluminous (about 1 cm³ or larger) brown pellet was formed, while the supernatant became clear once again.

Ammonia extract yielded the largest amount of solid residue without alkali titration and was chosen for SEM analysis. Dry ammonia water extract from shungite was prepared by mixing three parts of shungite rock and four parts of 25% ammonia water, and was allowed to infuse at room temperature for 24 ± 2 hours. The shungite ammonia water extract was concentrated in a rotary evaporator, and finally dry ammonia water extract was obtained by drying in a thermostat at 70° C.

Elemental and phase composition analysis

The elemental composition of raw shungite gravel was determined with an X-ray fluorescence (XRF) analyzer ZSX400 (Rigaku) using 10 mm diaphragms. The phase composition or raw rock and dried 25% ammonia extract of shungite was characterized by X-ray diffraction (XRD) method (Bragg-Brentano optics) using a SmartLab diffractometer (Rigaku), CuK α radiation (tube power 8.1 kW) and X-ray powder diffraction database ICDD PDF-2

(2015). The micrographs of the rock and the extracts were recorded using a scanning electron microscope TM3000 (Hitachi), which was equipped with an energy dispersive X-ray spectrometer SwiftED 3000 (Oxford Instruments) for determining the (local) chemical composition. A Spectrum BXII (Perkin Elmer) with ZnSe ATR (Attenuated Total Reflectance) was used for FTIR (Fourier-Transform Infrared Spectroscopy) measurements.

Microorganisms

The microorganisms used for determining the antibacterial activity were as follows: *Escherichia coli* ATCC 700336 (pyelonephritis strain), *Staphylococcus aureus* ATCC 43300 (methicillin-resistant strain), *Candida albicans* ATCC MYA-2876 (genome sequencing strain), *Pseudomonas aeruginosa* ATCC 27853 (quality control and susceptibility testing strain), *Streptococcus uberis* ATCC BAA-854/0140J (genome sequenced strain). Microbial suspensions were prepared with distilled water and a vortex device to obtain a visually opalescent suspension with turbidity comparable to the nephelometric 0.5 McFarland standard. Such opalescence refers to approximately 1.5×10^8 CFU/mL for bacteria.

Beer brewing with shungite

The beer kit MasterPint Czech Pilsner was used for 23 liters of homemade beer, plus 1 kg of grist with and without 1 kg of washed shungite gravel, with an original gravity of 1.040 (measured by hydrometer). The fermentation time was three weeks, the secondary fermentation time was one week. During the last two days, 50 g of Citra hops (*Humulus lupulus*) was added by dry hopping method.

Antimicrobial activity of shungite water extract

Distilled water was inoculated with cultures obtained from 24 ± 2 hours old, turbid cultures until a 0.5 McFarland microbial suspension was formed. 20 µL of the microbial suspension was added to 3 mL of distilled water that served as control, or to 3 mL of shungite infusion (experimental), and vortexed. After 24-hour incubation at room temperature in the dark and shaking, the samples and their 1:10, 1:100 and 1:1000 dilutions (volume 100 µL) were plated onto Mueller–Hinton agar (Oxoid) and incubated at 37 °C in the dark. 24 ± 2 hours later, the colonies were counted. The counts were compared with a two-tailed t-test and Fisher's exact test.

In order to measure the antibacterial activity of shungite water extract in the presence of abundant nutrients, the minimum inhibitory concentration (MIC) of 3:7 shungite water extract in Mueller–Hinton broth was determined by means of serial dilution. In addition, we compared ciprofloxacin MIC of *E. coli* and *P. aeruginosa* on solid Mueller–Hinton media, prepared as usual with distilled water with experimental 3:7 shungite water extract in order to detect any possible synergy between shungite water and fluoroquinolones.

RESULTS

Elemental and phase composition of shungite rock

Table 1 lists all the elements detected in raw shungite according to XRF. The main elements (concentration > 0.2 mass%) were the following: O, C, Si, Al, S, Fe, K,

 Table 1. List of elements and their concentrations detected in shungite

Concentration,	Concentration, Concentration, mass	
mass % of raw	of dried shungite water	
shungite	extract	
40.500	59.200	
28.100	0.517	
23.900	0.270	
2.080	4.980	
1.590	21.600	
1.440	6.950	
1.140	0.047	
0.550	2.160	
0.210	Not detected	
0.140	0.680	
0.080	0.008	
0.070	Trace amount	
0.060	Not detected	
0.020	1.140	
0.020	0.022	
0.008	0.020	
0.006	0.378	
0.005	0.006	
0.004	Trace amount	
0.002	1.630	
0.002	Not detected	
0.001	0.013	
Not detected	0.289	
Not detected	0.068	
Not detected	0.009	
Not detected	0.005	
	Concentration, mass % of raw shungite 40.500 28.100 23.900 2.080 1.590 1.440 1.140 0.550 0.210 0.140 0.050 0.020 0.020 0.020 0.020 0.020 0.020 0.020 0.002 0.008 0.006 0.005 0.004 0.005 0.004 0.002 0.002 0.001 Not detected Not detected Not detected	



Fig. 2. X-ray diffraction pattern of raw shungite mineral. Positions of the identified reflections are shown in the upper part of the figure for (1) quartz (α -SiO₂, PDF-2 file No. 46-1045), (2) muscovite (KAl₃Si₃O₁₀(OH)₂, PDF-2 file No. 84-1302), (3) pyrite (FeS₂, PDF-2 file No. 42-1340). arb. – arbitrary.

Ti and Mg. The XRD analysis showed (Fig. 2) that the main crystalline phase in the raw shungite was α -quartz (SiO₂, PDF-2 file No. 46-1045). The second phase was monoclinic muscovite and illite (KAl₃Si₃O₁₁, PDF-2 file No. 46-741 and 26-911), which is in good agreement with FTIR Si–O vibrations. The other trace phases were pyrite (FeS₂, PDF-2 file No. 42-1340) and probably also geikielite (MgTiO₃, PDF-2 file No. 06-0494). Non-crystalline carbon, the main constituent of the rock, was identified by an amorphous halo at a circular diffraction angle of 26.4°, corresponding to reflection 002 of graphite.

Chemical composition of shungite water extract

Figure 3 shows the FTIR spectrum of raw shungite. The strong absorption maximum at 1075 cm⁻¹ as well as at 885 cm⁻¹ belong to different Si–O bond vibrations. The very broad maximum of 3009 corresponds to the OH vibrations of acid. The weak signal at 2515 cm⁻¹ also supports the presence of acidic OH. At 1650 cm⁻¹, H₂O bending vibration was observed. There is also some contamination level of C–H stretching vibrations at 2892 and 2949 cm⁻¹.

Phase composition of ammonia water extract of shungite

XRD analysis showed (Fig. 4) that the main crystalline phase in the dried 25% ammonia water extract of shungite was mascagnite ($(NH_4)_2SO_4$ PDF-2 file No. 01-076-0579). The sample contained trace amounts of dolomite (PDF-2



Fig. 3. FTIR spectrum of raw shungite stone.

file No. 79-1344) and other unidentified crystalline phase(s) as well. The diffraction background around 20–30° was flat and did not show clear traces of an amorphous phase in the sample. The SEM micrographs of the ammonia water extract also agreed with the XRD outcome in describing the material as mainly crystalline.

Survival of microorganisms in shungite water

Candida albicans, Staphylococcus aureus and a commercially available *Saccharomyces cerevisiae* strain intended for beer brewing survived well in both distilled water and 3:7 shungite hot water extract without statistically significant changes or any overt trends. However, no colonies could be recovered after incubating *Echerichia coli*,



Fig. 4. X-ray diffraction pattern of shungite NH_3 extract. Positions of the identified reflections are shown in the upper part of the figure for mascagnite ($(NH_4)_2SO_4$). arb. – arbitrary.

Streptococcus uberis or Pseudomonas aeruginosa after 24-hour incubation in shungite water extract, as they had died (or were rendered unculturable; Table 2). The difference was highly significant for the first two bacteria (p = 0.0056 and p = 0.0031), while for *P. aeruginosa* the difference was statistically insignificant because of the high standard deviation of the negative control. The analysis was performed in seven separate instances. 1:10 shungite cold water extract did not inhibit the growth of microorganisms other than *E. coli* (no colonies had recovered after 120 minutes, the difference was statistically not significant, no data are shown).

Table 2. Survival of bacteria in distilled water (H₂O) versus shungite water (ShW)

	P. aeruginosa CFU/mL		S. uberis CFU/mL		<i>E. coli</i> CFU/mL	
	H ₂ O	ShW	H ₂ O	ShW	H ₂ O	ShW
1	188000	>100	29000	>100	76000	>100
2	20000	>100	78000	>100	370000	>100
3	63000	>100	102000	>100	189000	>100
4	6900	>100	57000	>100	63000	>100
5	9200	>100	11000	>100	500000	>100
6	2800	>100	44000	>100	500000	>100
7	30000	>100	58000	>100	404000	>100
Average	45700	0	54142	0	300285	0
stdev	65997	0	30218	0	189024	0
T-test p	0.116653965		0.003189906*		0.005666544*	

Antimicrobial activity of pH-neutralized shungite water

After overnight incubation, there was a triple mean of 103 000 CFU/mL in the distilled water, but no colonies in the shungite water or pH-neutralized shungite water. The test was performed in triplicates with approximately the same pH values (± 0.2). The results were similar (10⁵ CFU/mL in the distilled water, none in the shungite waters).

Determining the role of sulfite as an explanation for antibacterial activity

Sulfite content was measured in shungite water by means of iodometry in triplicates, using starch as an indicator. The ability of shungite water to reduce iodine was negligible or within the method's margin of error. However, instead of the characteristic dark blue color of the iodinestarch complex, a dark brown color was observed.

Effect of shungite rocks on beer brewing

The final gravity of beer brewed with shungite rocks (4% brew) was 1.007, which corresponds to 4.3% alcohol by volume. The final gravity of beer without shungite was 1.006, corresponding to 4.5% alcohol. The organoleptic properties of beer brewed with shungite gravel were subjectively indistinguishable from those of beer brewed without shungite. No other effects, including microbiological differences, were observed in the fermentation and maturation of beer brewed with plain water and shungite water.

DISCUSSION

According to XRF, the main atomic constituents of our shungite were oxygen, sulfur and carbon/nitrogen (the latter cannot be distinguished reliably). Calcium, as well as low concentrations of iron, sodium and magnesium were also found. This agrees with the XRD-determined iron sulfide (and ammonium sulfate of NH₃ extracts), with some additions – probably calcium carbonate/bicarbonate. According to the shungite classification (Ivankin 1987), based on the concentration of carbon, our raw shungite sample is class 3 shungite.

We attempted several solvents for extraction: room temperature water, hot water, 25% room temperature ammonia water, acetic acid, ethyl acetate and petrol ether. The ammonia water extract yielded the greatest amount of solid residue, and it was chosen for further characterization. The solid residue from the hot water extraction was much lower, while the solid residues from acetic acid, ethyl acetate and petrol ether extracts were negligible and could not be collected. In our pilot study (unpublished data), the hot water extract appeared microbiologically more potent than the cold water extract. We also determined previously that shungite water was highly microbicidal against E. coli, S. uberis and P. aeruginosa (after inoculation and 24-hour incubation, the distilled water contained approximately 10 000 to 100 000 CFU/mL bacteria, but the shungite waters were clear (Silver Türk unpublished data)). However, neither S. aureus nor C. albicans displayed such susceptibility to shungite water. So far, the ability of shungite filters to clean water containing E. coli has been reported (Khadartsev and Tuktamyshev 2002; Mosin and Ignatov 2012). Siwila and Brink (2019a) showed that the point-of-use water treatment technologies may often produce water free of pa- thogens with apparent removal of E. coli, but may not be affordable for the poorest groups in developing countries. A competing technology using geotextiles as low-cost drinking water treatment fabrics does not show a good effect in removing E. coli (Siwila and Brink 2019b).

Since shungite is acidic, we proposed that the low pH might be responsible for the antibacterial effect. Since pH-neutralized shungite water was antibacterial similarly to non-neutralized shungite water, the antibacterial properties of shungite water cannot be explained by low pH. Shungite water may contain aluminum, but in our sample, it was not a major component. Aluminum ions have been shown to be partly responsible for the antibacterial activity of certain clays (Lang 2013; Londono et al. 2017). Further analyses are necessary to clarify the role of aluminum (and other metal ions).

Since our shungite harbored 1.92% sulfur, we hypothesized that sulfite may be responsible for the antibacterial activity, in spite of the XRD analysis of the raw material suggesting the presence of sulfide and the XRD analysis of NH3 water extract suggesting the presence of sulfate. It is known (Lu et al. 2011) that sulfite is antibacterial against E. coli, more so than benzoic, sorbic and acetic acids. The formation of a dark brown color during titration of shungite water with iodine in the presence of starch suggests that unknown substances in shungite water modify the formation of iodine-starch complexes. Given that the shungite water vapor contains yellowish or golden crystals, we considered the possibility that it may contain either elemental iron sulfide (as suggested by XRD of raw shungite), derived sulfates (as suggested by XRD of ammonia extract) or possibly elemental sulfur, which are not likely explanations for antibacterial effects. It seems unlikely that the antibacterial activity of shungite water is due to the presence of sulfite ions.

Unlike the antibacterial properties of shungite water in the context of shungite water inoculated with bacteria without the presence of nutrients, shungite water extract did not display antimicrobial properties in the serial dilution test of Mueller–Hinton broth, even at 50% concentration of shungite water. Therefore, its antibacterial property is not comparable to that of antibiotics.

The susceptibility to ciprofloxacin on Mueller–Hinton agar prepared with shungite water was compared with the susceptibility to ciprofloxacin on standard Mueller–Hinton agar. It was expected that the MIC would decrease for *E. coli*. Surprisingly, it was found that shungite water lowered the MIC of *P. aeruginosa* instead (0.38 μ g/mL vs 1 μ g/mL).

The uniform lack of bacteria in shungite water evident in our study was not present in our pilot study (unpublished data) with more dilute preparations, such as 1:10, which yielded very variable results. Hence, we decided to use relatively potent 3:7 extracts for this study.

In an earlier analysis (Efremova 2006), Candida strain 10K and Pseudomonas strain 51K were cultured in the presence of shungite and oil in the context of oil bioremediation (Efremova 2006). In our study, only Candida species survived well in shungite water, while Pseudomonas died or became unculturable in the presence of shungite water extract. The latter difference may be caused by the relative potency of our shungite water extract due to the difference between shungites (our shungite was Karelian in origin, while the cited study used shungite from Kazakhstan), or the difference between Pseudomonas species (we used P. aeruginosa strain ATCC 27853, whereas the cited study used Pseudomonas sp. strain 51K). While our pilot study suggested that the effect of shungite water is concentration-dependent, the possibility that the susceptibility of shungite water is strain-specific cannot be excluded. In addition, it cannot be excluded that shungite rock actually has practical water purifying properties by mechanisms of adsorption (as described previously) or harboring a biofilm that is antagonistic to certain undesirable water contaminants such as Escherichia coli.

Finally, we tested beer brewing with shungite extract to test possible food industry applications. Brewing on 4% shungite gravel did not interfere with the brewing process.

CONCLUSIONS

Shungite water has weak antibacterial properties. 3:7 shungite hot water extract is not conducive to the 24-hour survival of *E. coli, P. aeruginosa* and *S. uberis*. Shungite water is conducive to the survival of *S. aureus, C. albicans* and *S. cerevisiae*. Mueller–Hinton media prepared with shungite water is not antibacterial. The antibacterial properties of shungite water cannot be explained by either acidic pH or sulfite content. Beer prepared with 4% shungite gravel was organoleptically similar to the control beer, albeit with a slightly lower alcohol content.

ACKNOWLEDGEMENTS

This study was supported by the Estonian Research Council and the Estonian Ministry of Education and Research (grants Nos IUT34-19, IUT20-17, IUT2-24, IUT34-19, KOGU-HUMB) as well as Enterprise Estonia (grant No. EU48695). The authors are thankful to Peeter Ritslaid for XRF measurements. The publication costs of this article were covered by the Estonian Academy of Sciences.

REFERENCES

- Baati, T., Bourasset, F., Gharbi, N., Njim, L., Abderrabba, M., Kerkeni A., Szwarc, H. and Moussa, F. 2012. The prolongation of the lifespan of rats by repeated oral administration of [60] fullerene. *Biomaterials*, 33(19), 4936– 4946.
- Ballantyne, E. N. 1930. On certain factors influencing the survival of bacteria in water and in saline solutions. *J. Bacteriol.*, 19(5), 303–320.
- Buseck, P. R. 2002. Geological fullerenes: review and analysis. *Earth Planet. Sci. Lett.*, **203**(3–4), 781–792.
- Charykova, M. V., Bornyakova, I. I., Polekhovskii, Y. S., Charykov, N. A., Kustova, E. V. and Arapov, O.V. 2006. Chemical composition of extracts from shungite and "shungite water". *Russ. J. Appl. Chem.*, **79**(1), 29–33.
- Dallakyan, G. A., Pogosyan, S. I. and Ipatova, V. I. 2018. The combined effect of shungite and heavy metals on the growth of microalgae population. *Inland Water Biol.*, 11(1), 103–107.
- Efremova, S. V. 2006. Water treatment with a shungite sorbent and biosorbents on its base. *Russ. J. Appl. Chem.*, **79**(3), 397–402.
- Grohn, K. J., Moyer, B. S., Wortel, W. C., Fisher, C. M., Lumen, E., Bianchi, A. H. et al. 2021. C₆₀ in olive oil causes lightdependent toxicity and does not extend lifespan in mice. *GeroScience*, 43(2), 579–591.
- Ivankin, P. F., Galdobina, L. P. and Kalinin, Y. K. 1987. Shungites: origin and classification of a new carbon mineral resource. *Int. Geol. Rev.*, 29(10), 1208–1214.
- Khadartsev, A. A. and Tuktamyshev, I. S. 2002. Шунгиты в медицинских технологиях (Shungites in medical technology). Вестник новых медицинских технологий, 9(2), 83–86 (in Russian).
- Kovalevski, V. V. and Moshnikov, I. A. 2016. TEM study of structure of graphene layers in shungite carbon. *Nanosyst.*: *Phys., Chem., Math.*, 7(1), 210–213.
- Lang U. 2013. Minimal inhibitory concentration of natural vinegar and of aluminium acetate-tartrate solution. *Otol. Neurotol.*, 34(5), 795–797.
- Liu, S., Zeng, T. H., Hofmann, M., Burcombe, E., Wie, J., Jiang, R. et al. 2011. Antibacterial activity of graphite, graphite oxide, graphene oxide, and reduced graphene oxide: membrane and oxidative stress. ACS Nano, 5(9), 6971– 6980.
- Londono, S. C., Hartnett, H. E. and Williams, L. B. 2017. Antibacterial activity of aluminum in clay from the Colombian Amazon. *Environ. Sci. Technol.*, **51**(4), 2401–2408.

- Lu, H. J., Breidt, F., Pérez-Díaz, I. M. and Osborne, J. A. 2011. Antimicrobial effects of weak acids on the survival of *Escherichia coli* O157:H7 under anaerobic conditions. J. *Food Prot.*, 74(6), 893–898.
- Lyn'kov, L. M., Borbot'ko, T. V. and Krishtopova, E. A. 2009. Radio-absorbing properties of nickel-containing schungite powder. *Tech. Phys. Lett.*, 35(5), 410–411.
- Mosin, O. V. and Ignatov, I. 2012. Состав и структурные свойства природного фуллеренсодержащего минерала шунгита (The composition and structural properties of fullerene natural mineral shungite). *Наноинженерия*, 18(12), 17–24 (in Russian).
- Mosin, O. V. and Ignatov, I. 2013. The structure and composition of natural carbonaceous fullerene containing mineral shungite. *Int. J. Adv. Sci. Tech. Res.*, 6(3), 9–21.
- Rozhkov, S. P. and Goryunov, A. S. 2013. Interaction of shungite carbon nanoparticles with blood protein and cell components. *Russ. J. Gen. Chem.*, 83(13), 2585–2595.
- Sajo, M. E. J., Kim, C.-S., Kim, S.-K., Shim, K. Y., Kang, T.-Y. and Lee, K.-J. 2017. Antioxidant and anti-inflammatory effects of shungite against ultraviolet B irradiation-induced skin damage in hairless mice. *Oxid. Med. Cell Longev.*, 2017, ID7340143.
- Santos, V. G., Fasciotti, M., Pudenzi, M. A., Klitzke, C. F., Nascimento, H. L., Pereira, R. C. L. et al. 2016. Fullerenes in asphaltenes and other carbonaceous materials: natural constituents or laser artifacts. *Analyst*, **141**(9), 2767–2773.

- Serrano, C., Romero, M., Alou, L., Sevillano, D., Corvillo, I., Armijo, F. and Maraver, F. 2012. Survival of human pathogenic bacteria in different types of natural mineral water. J. Water Health, 10(3), 400–405.
- Sheka, E. F. and Rozhkova, N. N. 2014. Shungite as the natural pantry of nanoscale reduced graphene oxide. *Int. J. Smart Nano Mater.*, 5(1), 1–16.
- Sheka, E. F., Natkaniec, I., Rozhkova, N. N. and Holderna-Natkaniec, K. 2014. Neutron scattering study of reduced graphene oxide of natural origin. *JETP Lett.*, 99(11), 650– 655.
- Siwila, S. and Brink, I. C. 2019a. Comparison of five point-ofuse drinking water technologies using a specialized comparison framework. J. Water Health, 17(4), 568–586.
- Siwila, S. and Brink, I. C. 2019b. Low cost drinking water treatment using nonwoven engineered and woven cloth fabrics. J. Water Health, 17(1), 98–112.
- Skrypnik, L., Babich, O., Sukhikh, S., Shishko, O., Ivanova, S., Mozhei, O. et al. 2021. A study of the antioxidant, cytotoxic activity and adsorption properties of Karelian shungite by physicochemical methods. *Antioxidants (Basel)*, **10**(7), 1121.
- Tyutyunnik, N. N., Unzhakov, A. R., Uzenbaeva L. B., Golubeva, A. G., Ilykha, V. A., Kalinina, S. N. et al. 2009. Influence of shungite carbon nanoparticles on the physiological condition of dark brown mink (Neovision Vision SCHR. 1777). *Scientifur*, 33(3), 88.

Šungiidivee mikrobioloogilised ja keemilised omadused

Silver Türk, Tarmo Tamm, Hugo Mändar, Ain Raal, Peeter Laurson ja Uno Mäeorg

Šungiidifiltreid on kasutatud *Escherichia coli* eemaldamiseks veest. Šungiidivee toimemehhanism ja toimespekter pole teada. Käesolev uuring kirjeldab šungiiti ja selle vesiekstrakti röntgendifraktsiooni, röntgenfluorestsentsi ja jodomeetria abil. Šungiidi vesiekstrakti kuivjääk sisaldas suhteliselt vähe süsinikku (28,1% kivimis vs 0,5% kuivjäägis), räni (23,9% vs 0,3%) ja kaaliumi (1,14% vs 0,05%) ning suhteliselt palju väävlit (1,6% vs 21,6%), rauda (1,4% vs 10%), alumiiniumi (2,1% vs 5%) ja niklit (0,02% vs 1,14%).

Selgitati välja šungiidivee mõju *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Candida albicans, Streptococcus uberis* ja *Saccharomyces cerevisiae* suhtes. *Escherichia coli, Pseudomonas aeruginosa* ja *Streptococcus uberis* ei olnud 24h inkubatsiooni 3:7 ekstraktis välja külvatavad. *Staphylococcus aureus, Candida albicans* ja *Saccharomyces cerevisiae* elasid inkubatsiooni šungiidivees üle võrreldavalt destilleeritud veega. Vee pH neutraliseerimine ei kaotanud bakteritsiidset toimet. Samas, toitainete juuresolekul ei ilmnenud šungiidiveel bakterivastast mõju.