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The Effects on Some Parameters of Microbiological and Physicochemical Research of Zeolite and Zeolite Filtered Water from Rhodope Mountains, Bulgaria

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Abstract: The physicochemical composition and antibacterial effect of aqueous zeolite infusion (spring zeolite water Sevtopolis with zeolite from deposit Beli plast, Bulgaria), obtained for 12 and 36 hours, were tested. Ordinance No. 9/2001, Official State Gazette, issue 30, and Decree No.178/23.07.2004 regarding the quality of water intended for drinking and household purposes were applied to study the physicochemical composition. *Staphylococcus aureus*-ATCC and TSA-MRSA, and *Escherichia coli* ATCC were used in the studies. Both tested zeolite waters reduced the amounts of viable *E. coli* and *S. aureus* cells even when they were in high concentrations (10^6 cells/mL). The effect of the 36-hour infusion was better, under the influence of which, after 60 minutes, the number of live bacteria of both tested species decreased by almost half compared to the initial amounts. Slightly higher sensitivity to two zeolite waters was shown by *E. coli*, whose cells were reduced to about 35% after two hours of exposure to both zeolite waters tested. However, about 20% of the cells of the tested bacteria survived even after 96 hours of exposure to these waters. Only in *E. coli* no growth was found after 96 hours of exposure to 12 hours of zeolite water. These results show the effectiveness of zeolite for water purification from Gram-positive and Gram-negative bacteria, as well as prospects for using zeolite water as a prophylactic and auxiliary treatment for bacterial infections.

Keywords: zeolite water, Beli plast zeolite deposit, antibacterial activity, physicochemical parameters

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INTRODUCTION

Scientific research shows that in areas where there is zeolite, the mineral, with its antimicrobial properties and physicochemical composition of zeolite water, can benefit public health and longevity (1-5). Zeolite is used in filtration systems for water purification (6,7). In Southern Bulgaria, there are six zeolite deposits in the Rhodope Mountains. They are located in the northeastern Rhodopes. These are Beli Plast, Most, Gorna Krepost, Golobradovo, Lyaskovets and Beliat Bair. Clinoptilolite and modernite are the primary zeolite materials in them. In this area, the water passes through zeolite layers.

Zeolites are aluminosilicate members of the family of microporous solids, known as "molecular sieves" and consist mainly of silicon, aluminum, and others. They have a porous structure that can hold a wide variety of cations, such as Na^+, K^+, Ca^{2+}, Mn^{2+}, and others. These positive ions are relatively weakly bound and can be easily replaced by others in contact solution. Some of the most common mineral zeolites are analzyme, chabazite, stilbit, clinoptilolite, hyulandite, natrolite and phillipsite. These cations exchanged zeolites have different acidity and catalyze several acids catalyzes (8, 9). Because zeolites are microporous aluminosilicate minerals, they are often used as adsorbents and catalysts. Zeolite is a non-toxic, three-dimensionally porous, crystalline, hydrated aluminosilicate with natural adsorbent and ion exchange properties, which removes harmful microbes as well as dispersed insoluble and soluble toxins from drinking water. Zeolite has been used since the Maya civilization for water purification and decontamination (10, 11). Nowadays, creating a simple flow filter made of zeolite-cotton packing in a tube (ZCT) provides a solution as an inexpensive device for removing heavy metal ions from contaminated water with high adsorption efficiency. After flowing through the setup packed with 10 g of zeolite-cotton, 65 mL 1000 ppm Cu^{2+} solution is purified down to its safety limit (<1 ppm) (12). ZCT can be used for disinfection by introducing Ag-exchanging zeolite-cotton for silver ion exchanging by incorporating them into a commercial dental adhesive and investigating the inhibition of biofilm formation at the tooth-restoration margin (13).

Zeolites for industrial use are also produced synthetically. This is done by heating aqueous alumina and silica solutions with sodium hydroxide. Equivalent reagents include sodium aluminate and sodium silicate. Synthetic zeolites have some key advantages over their natural counterparts. Synthetic materials are produced in a homogeneous, phase pure state. It is also possible to obtain zeolite structures that do not occur in nature. As the main raw materials used for producing zeolites are silicon and aluminum, one of the most common mineral components on earth, the potential for supplying zeolites is practically unlimited (14). This allows their widespread use as ion exchanger means for domestic and commercial water purification, softening, and more. In chemistry, zeolites are used to separate molecules (only molecules of certain sizes and shapes can be omitted) and as molecule traps so that they can be analyzed. Zeolites are also widely used as catalysts and sorbents. Their well-defined pore structure and regulated acidity make them highly active in a wide variety of reactions (15). Their application as filter additives in aquariums for the adsorption of ammonia and other nitrogen compounds is also very useful. Due to the adsorption properties of zeolites for calcium, they may effectively reduce calcium in hard water (1). Recently, studies have been presented on a new type of zeolite, finding that it helps increase eggplant yields by supplying potassium, calcium, and iron ions to the soil (16). This zeolite has a good content of nutrients necessary for plants and physical properties for water retention, making it an important clay mineral for agricultural and industrial purposes.

The research and development of many biochemical and biomedical applications of zeolites, particularly the naturally occurring species heulandite, clinoptilolite, and chabazite, continues. (18). In the present work, the authors performed physicochemically and *in vitro* studies to test the antibacterial properties of zeolite waters with zeolite from deposit Beli Plast (Figure 1) (17-20).

Figure 1: Zeolite, Beli plast, Bulgaria. Natural deposit,"Zeolitni gabi".

The water used for the research was spring water "Sevtopolis" (21). It was obtained via filtration with zeolite from Beli plast, Bulgaria. The physicochemical parameters of that zeolite and the water that filtered through it, as well as their antimicrobial effects, have not yet been studied. We have selected some of the most important conditionally pathogenic Gram-positive (*S. aureus*) and Gram-negative (*E. coli*) bacteria of great importance in the infectious pathology of animals and humans. The effects on them would be indicative of other similar microorganisms.

The study aims to show with microbiological and physicochemical parameters that zeolite has the potential for important positive effects on public health and environmental cleanliness in the areas where it is used.

MATERIALS AND METHODS

Zeolite

The effect of aqueous infusions of zeolite water was obtained by soaking 50 g of zeolite from Beli plast (Bulgaria) in the form of granules (0.5 cm in diameter) in 500 mL of zeolite water for 12 and 36 hours at room temperature 20 ± 0.5 °C.

Chemical composition of Zeolite from Beli plast (Bulgaria)

Table 1 illustrates the chemical composition of the tested zeolite in % (w/w).

Table 1. The chemical composition of zeolite from Beli plast (Bulgaria) in % (w/w)

No.	Chemical component	Content, % (w/w)
1	SiO_2	66.6
2	Al_2O_3	11.41
3	Fe_2O_3	0.8
4	MgO	0.06
5	TiO_2	0.15
5	CaO	2.80
6	Na_2O	0.22
7	K_2O	2.9

Physical indicators. The physical and chemical indicators are pH, oxidation-reduction potential (ORP), and temperature of zeolite waters were measured using Manual multi-parameter analyzer Consort C1010 (Consort bvba, Belgium) for pH, mV, and temperature measurement).

Microorganisms. Suspensions with a concentration of 10^7 cells.mL^{-1} of two strains of Staphylococcus aureus - ATCC-6538 and TSA-MRSA, as well as Escherichia coli ATCC-8739 were used in the studies. The suspensions were prepared in sterile saline by the Mc Ferland standard optical method.

Nutrient media.

Mueller Hinton agar (BUL BIO NCIPD - Sofia) was used to obtain 24-hour cultures of the tested strains, as well as solid selective media (Antisel - Sharlau Chemie SA, Spain) to determine the effect of the tested zeolite waters for antimicrobial activity: Eosin Methylene Blue agar for *E. coli* and Chapman Stone agar for *S. aureus*.

Methods for determination of microbiological indicators from Bulgarian State Standard

1. Methods for evaluating microbiological indicators according to Ordinance No. 9/2001, Official State Gazette, issue 30, and decree No. 178/23.07.2004 about the quality of water intended for drinking purposes.
2. Method for determination of *Escherichia coli* and coliform bacteria – BDSEN ISO 9308-1: 2004;
3. Method for determination of enterococci – BDS EN ISO 7899-2;
4. Method for determination of sulfite reducing spore anaerobes – BDS EN 26461-2: 2004;
5. Method for determination of the total number of aerobic and facultative anaerobic bacteria – BDS EN ISO 6222: 2002;
6. Method for determination of *Pseudomonas aeruginosa* – BDS EN ISO 16266: 2008.
7. Determination of coli – titre by fermentation method – Ginchev's method

Determination of coli – bacteria over Endo's medium – membrane method.

8. Determination of sulfite reducing anaerobic bacteria (*Clostridium perfringens*) – membrane method.

The number of the isolated bacteria was presented in colony-forming units per mL (CFU.mL^{-1}) of the tested waters (22).

Methods for physicochemical analysis.

The physicochemical analysis was performed according to Ordinance No. 9/2001, Official State Gazette, issue 30, and Decree No. 178/23.07.2004 regarding the quality of water intended for drinking and household purposes. The following methods are applied (Ordinance No. 9/2001) (22):

The determination of color was according to Rublyovska Scale – the method by Bulgarian State Standard (BDS) 8451: 1977;

Method for determining smell at 20°C — method BDS 8451: 1977 technical device – glass mercury thermometer, conditions No. 21;

Method for determination of turbidity - EN ISO 7027, with technical device turbidimeter type TURB 355 IR ID No 200807088;

Method for determination of pH – BDS 3424: 1981, technical device pH meter type UB10 ID NoUB10128148;

Method for determination of oxidisability – BDS 3413: 1981;

Method for determination of chlorides – BDS 3414: 1980;

Method for determination of nitrates – Validated Laboratory Method (VLM) – NO_3 – No. 2, technical device photometer "NOVA 60 A" ID No. 08450505;

Method for determination of nitrites – VLM NO_2 – No. 3, technical device photometer "NOVA 60 A" ID No. 08450505;

Method for determination of ammonium ions – VLM – NH_4 – No. 1, technical device photometer "NOVA 60 A" ID No. 08450505;

Method for determination of general hardness – BDS ISO 6058;

Method for determination of sulfates – VLM – SO_4 – No. 4, technical device photometer "NOVA 60 A" ID No. 08450505;

Method for determination of calcium – BDS ISO 6058;

Method for determination of magnesium – BDS 7211: 1982;

Method for determination of phosphates – VLM - PO_4 – No. 5, technical device photometer "NOVA 60 A" ID No 08450505;

Method for determination of manganese – VLM – Mn – No. 7, technical device photometer "NOVA 60 A" ID № 08450505;

Method for determination of iron – VLM – Fe – No. 6, technical device photometer "NOVA 60 A" ID No. 08450505;

Method for determination of fluorides – VLM – F – No. 8, technical device photometer "NOVA 60 A" ID No. 08450505;

Method for determining electrical conductivity – BDS EN 27888, technical device – conductivity meter inoLab cond 720 ID No 11081137.

Experimental staging

To 4.5 mL of a 12-hour as well as 36-hour zeolite water, 0.5 mL of a suspension of the corresponding bacterial strain was added at a concentration of 10^7 cells. mL^{-1}, reaching a final concentration of 10^6 cells/mL for each species and strain of microorganisms used. The following controls were placed - 12 and 36-hour zeolite water without microorganisms, as well as bacteria without zeolite water. After different time intervals for exposure to zeolite waters (15 min, 30 min, 60 min, 120 min, 24 h, and 96 h), cultures were made from each of the samples on Eosin Methylene Blue selective agar medium for the Gram-negative bacteria and Chapman Stone agar for S. aureus in order to determine the antimicrobial activity of the tested waters against E. coli and S. aureus. After culturing at 37 °C for 18-24, the growth of the tested bacteria was reported, as well as that of the set controls. Colonies formed were counted, and the results were calculated in colony-forming units/mL ($CFU.mL^{-1}$) as a percentage of the growth of untreated controls of the strains, which were considered 100 %.

Statistical analysis. The results were processed mathematically, and the arithmetic mean (AV) and standard deviation (SD) were found. Student's t-test analysis for independent samples was applied to test the statistical dependence and reliability of the results. The significance of the results was defined at a significance level of $P<0.05$. Microsoft®Office Professional Plus Excel 2013 (15.0.4569.15060) was used for the calculations.

RESULTS AND DISCUSSION

Physicochemical analysis of spring water with zeolite filtration "Sevtopolis"

Table 2 illustrates physicochemical parameters of spring water with zeolite filtration "Sevtopolis" according to Ordinance No. 9/2001. The research is with the document (Laboratory Alimenti Omnilab, Plovdiv, Bulgaria, No. 10838, 22.02.2021)

Table 2. Physicochemical parameters of spring water with zeolite filtration "Sevtopolis"

Controlled parameter	Measuring unit	Maximum Limit Value	Spring water with zeolite filtration "Sevtopolis"
1. pH	pH values	≥ 6,5 and ≤ 9,5	6.91±0.1
2. Electrical conductivity	$\mu S.L^{-1}$	2000	278 ±8.3
3. Color	Chromaticity Values	Acceptable	Acceptable
4. Turbidity	FNU	Acceptable	Acceptable
5. Odor	force	Acceptable	Acceptable
6. Calcium (Ca^{2+})	mg. L^{-1}	150	34.8±3.5
7. Magnesium (Mg^{2+})	mg. L^{-1}	80	8.26±0.83
8. Zinc (Zn^{2+})	mg. L^{-1}	4.0	<2.0
9. Manganese (Mn^{2+})	µg. L^{-1}	50	<10.0
10. Sodium (Na^{+})	mg. L^{-1}	200	13.85±1.4
11. Potassium (K^{+})	mg. L^{-1}	-	3.57±0.36
12. Nitrites (NO_2^{-})	mg. L^{-1}	0.5	<0.05
13. Nitrates (NO_3^{-})	mg. L^{-1}	50	11.80±0.12
14. Sulfates (SO_4^{2-})	mg. L^{-1}	250	41.40±0.41
15. Ammonium (NH_4^{+})	mg. L^{-1}	0.5	<0.013
16. Chlorine (Cl^{-})	mg. L^{-1}	0.3-0.4	0.018
17. Iron (Fe)	µg. L^{-1}	200	<5.0
18. Selenium (Se)	µg. L^{-1}	10	<2.0
19. Antimony (Sb)	mg. L^{-1}	5	<2.7
20. Aluminum (Al)	µg. L^{-1}	200	<40.00
21. Lead (Pb)	µg. L^{-1}	10	<2.0
22. Boron (B)	mg. L^{-1}	1	<0.02
23. Arsenic (As)	µg. L^{-1}	10	<2.0
24. Chromium (Cr)	µg. L^{-1}	50	<10.0
25. Cadmium (Cd)	µg. L^{-1}	5	<1.0
26. Copper (Cu)	mg. L^{-1}	2	<0.08
27. Nickel (Ni)	µg. L^{-1}	20	<4.0
28. Mercury (Hg)	µg. L^{-1}	1	<0.5

The physical parameters of pH and ORP of the studied zeolite waters at room temperature 20° C are presented in Table 3.

Table 3. Physical indicators of the tested zeolite water

Zeolite water	pH	ORP (mV)
12 h	6.76 ± 0,1	0.7 ± 2.52
36 h	6.5 ± 0,04	32.3 ± 0.58

As can be seen from the data in the table, the zeolite water obtained for 12 and 36 hours have a slightly acid reaction and a neutral to slightly high ORP. A lower pH and a higher ORP indicated the water obtained for a longer time (36 h) compared to that obtained for 12 h. The differences between both waters for pH were statistically significant ($P < 0.05$), as well as for ORP ($P < 0.01$).

The results of the studies performed to determine the sensitivity of the used strains of *E. coli* and *S. aureus* to zeolite waters, tested at a final concentration of 10^6 cells.mL^{-1} by the suspension method, are presented in Table 4. Bacterial growth was defined as the percentage compared to growth of zeolite-free bacterial controls, which were considered 100%.

The data indicated that the two tested aqueous infusions of zeolite exhibited an antimicrobial effect, although it was not strongly pronounced. After 30 minutes of exposure, they reduced the amount of viable *E. coli* and *S. aureus* cells even when they were in high concentrations (10^6 cells.mL^{-1}). The effect of the 36-hour infusion was better, under the influence of which, after 60 minutes, the number of live bacteria of the two tested species decreased by almost half compared to the initial amounts. Slightly higher sensitivity to the zeolite waters was shown by *E. coli*, whose cells were reduced to about 35% after two hours of exposure to both zeolite waters tested. *S. aureus* cells were reduced to a lesser extent under the action of 12 h zeolite water. However, about 20% of the cells of the tested bacteria survived even after 96 hours of exposure to these waters. Only in *E. coli* no growth was found after 96 hours of exposure to 12 h zeolite water.

Table 4 and figures 3 and 4 illustrate the antimicrobial effect of 12 h zeolite water and 36 h zeolite water against *S. aureus,* and *E. coli* strains in suspensions with a density 10^6 cells. mL^{-1}

Table 4. Antimicrobial effect of 12 h zeolite water and 36 h zeolite water against *S. aureus* and *E. coli* strains in suspensions with a density 10^6 cells. mL^{-1}

Time of action	**Growth of the strains (% of CFU.mL^{-1}) after different intervals of exposure to zeolite waters**			
	S. aureus		***E. coli***	
	12 h	**36 h**	**12 h**	**36 h**
15 min	100.0 ±10.0	100.0 ±10.0	100.0 ±10.0	100.0 ±10.0
30 min	77.5 ±4.3	80.0 ±0.0	82.5 ±3.5	87.5 ±2.5
60 min	62.5 ±8.3	55.0 ±5.0	55.0 ±5.0	52.5 ±2.5
120 min	50.0 ±10.0	35.0 ±5.0	35.0 ±5.0	32.5 ±2.5
24 h	50.0 ±10.0	35.0 ±5.0	32.5 ±2.5	30.0 ±0.0
96 h	22.0 ±0.0	20.0 ±10.0	0	18.0 ±10.0
Untreated controls	100.0 ±10.0	100.0 ±10.0	100.0 ±10.0	100.0 ±10.0
Control without bacteria	0	0	0	0

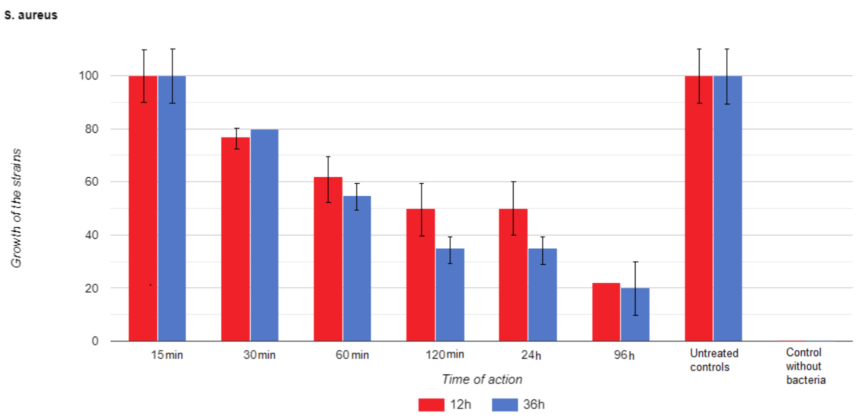

Figure 2: Antimicrobial effect of 12 h zeolite water and 36 h zeolite water against *S. aureus* and strains in suspensions with a density 10^6 cells. mL^{-1}

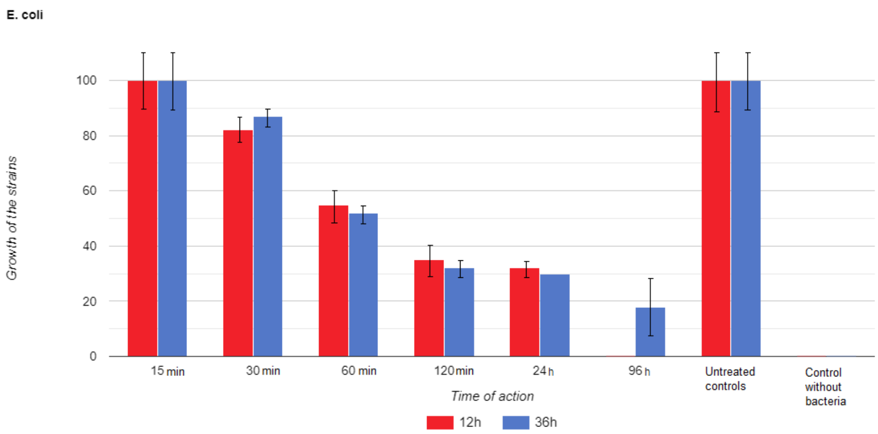

Figure 3: Antimicrobial effect of 12 h zeolite water and 36 h zeolite water against *E. coli* strains in suspensions with a density 10^6 cells. mL^{-1}

Microbiological research of this standard was performed by Bulgarian authors (23-26). Data from many of them show that silver ions enhance the antimicrobial properties of zeolite (27, 28). The research was performed with *in vitro* antimicrobial effect of fabric softeners containing silver zeolite on *S. aureus, Pseudomonas aeruginosa,* and *Candida albicans* (29). The studies demonstrated the inhibitory action of silver-zeolite-containing samples on *S. aureus*, including MRSA, as well as on *C. albicans* (30). In studies of the Mexican silver zeolite mineral clinoptilolite - heulandite demonstrated that it eliminated pathogenic microorganisms *E. coli* and *Streptococcus faecalis* from the water after 2 hours of contact (31). Silver ZCT exhibits antibacterial properties by completely purifying *E. coli*-contaminated water from the bacterial cells as it flows through the device (7). The zeolite water filter is a sustainable, natural solution for purifying drinking water and wastewater (32). Zeolite filters have many pores, so they not only trap particles between the grains but absorb them into their pores. This is partly done by zeolite minerals' ability to cation exchange, whereby they absorb positive ions from water (i.e., dissolved metals, sodium, ammonia, etc.). Also, due to its high pore density, zeolite has a highly effective surface and can trap high concentrations of contaminants before the filter needs to be cleaned. For these reasons, it can be used successfully to treat drinking water, wastewater, radioactive water, freshwater aquaculture, and others (33). The zeolite formulas can be combined with various materials used to manufacture medical devices, surfaces, textiles, or household items where antimicrobial properties are required.

Our research also shows that zeolite can be used to reduce the bacterial water content even when it is significant.

CONCLUSIONS

By examining the physicochemical and antimicrobial properties of zeolite, the study shows that it positively affects public health and environmental cleanliness in the areas where it is distributed.

The tested zeolite water, obtained for 12 and 36 hours, reduced the amount of viable *E. coli* and *S. aureus* cells even when they were in high concentration (10^6 cells/mL). *E. coli* showed a higher sensitivity to these waters' action than *S. aureus*.

REFERENCES

1. Tinteri C, Potenza M, Rizzetto R. Antimicrobial efficacy and longevity of silver+ zeolite incorporating preinsulated ducts installed in real healthcare settings. J prev med hyg. 2012;53(4):177–80.

2. Ernst WG. Overview of naturally occurring Earth materials and human health concerns. Journal of Asian Earth Sciences [Internet]. 2012 Oct [cited 2022 Jul 12];59:108–26. Available from: <URL>.

3. Ignatov I, Mosin O, Bauer E. Carbonaceous Fullerene Containing Mineral Shungite and Aluminosilicate Mineral Zeolite. Mathematical Model and Practical Application of Water Solution of Shungite and Zeolite. Journal of Medicine, Physiology and Biophysics. 2014;4:27–44.

4. Ibrahim SS, Salem NY. Insecticidal efficacy of nano zeolite against Tribolium confusum (Col., Tenebrionidae) and Callosobruchus maculatus (Col., Bruchidae). Bulletin of the National Research Centre. 2019;43(1):1–8.

5. Peixoto P, Guedes JF, Rombi E, Fonseca AM, Aguiar CA, Neves IC. Metal Ion–Zeolite Materials against Resistant Bacteria, MRSA. Ind Eng Chem Res [Internet]. 2021 Sep 8 [cited 2022 Jul 12];60(35):12883–92. Available from: <URL>.

6. Luo J, Lindsey S. The use of pine bark and natural zeolite as biofilter media to remove animal rendering process odours. Bioresource Technology [Internet]. 2006 Sep [cited 2022 Jul 12];97(13):1461–9. Available from: <URL>.

7. Chen W, Acharya D, Liu Z, Yi X, Xiao Y, Tang X, et al. Mechanistic insights of selective syngas conversion over Zn grafted on ZSM-5 zeolite. Catal Sci Technol [Internet]. 2020 [cited 2022 Jul 12];10(24):8173–81. Available from: <URL>.

8. Marakatti VS, Rao PVC, Choudary NV, Ganesh GS, Shah G, Maradur SP, et al. Influence of Alkaline Earth Cation Exchanged X-Zeolites Towards <I>Ortho</I>-Selectivity in Alkylation of Aromatics: Hard-Soft-Acid-Base Concept. adv porous mat [Internet]. 2014 Dec 1 [cited 2022 Jul 12];2(4):221–9. Available from: <URL>.

9. Marakatti VS, Halgeri AB. Metal ion-exchanged zeolites as highly active solid acid catalysts for the green synthesis of glycerol carbonate from glycerol. RSC Adv [Internet]. 2015 [cited 2022 Jul 12];5(19):14286–93. Available from: <URL>.

10. Flanigen EM. Chapter 2 Zeolites and Molecular Sieves an Historical Perspective. In: Studies in Surface Science and Catalysis [Internet]. Elsevier; 1991 [cited 2022 Jul 12]. p. 13–34. Available from: <URL>.

11. Tankersley KB, Dunning NP, Carr C, Lentz DL, Scarborough VL. Zeolite water purification at Tikal, an ancient Maya city in Guatemala. Sci Rep [Internet]. 2020 Dec [cited 2022 Jul 12];10(1):18021. Available from: <URL>

12. Chen X, Yu L, Zou S, Xiao L, Fan J. Zeolite Cotton in Tube: A Simple Robust Household Water Treatment Filter for Heavy Metal Removal. Sci Rep [Internet]. 2020 Dec [cited 2022 Jul 12];10(1):4719. Available from: <URL>.

13. Li W, Qi M, Sun X, Chi M, Wan Y, Zheng X, et al. Novel dental adhesive containing silver exchanged EMT zeolites against cariogenic biofilms to combat dental caries. Microporous and Mesoporous Materials [Internet]. 2020 Jun [cited 2022 Jul 12];299:110113. Available from: <URL>.

14. Rollmann L, Valyocsik E, Shannon R. Zeolite Molecular Sieves. In: Murphy D, Interrante L, editors. Inorganic

Syntheses. New York: Wiley and Sons, Inc.; 1995. p. 227–34.

15. Bhatia S. Zeolite Catalysts: Principles and applications. Boca Raton: CRC Press; 1989. 304 p.

16. Yasir A, Janabi N. Discovered a new type of zeolite and tested on some chemical properties of soil and plant yield. Plant Archives. 2020;Supp 1:1978–82.

17. Djurova E, Stefanova I, Gradev G. Geological, mineralogical and ion exchange characteristics of zeolite rocks from Bulgaria. Journal of Radioanalytical and Nuclear Chemistry, Articles [Internet]. 1989 Apr [cited 2022 Jul 12];130(2):425–32. Available from: <URL>.

18. Petrova N, Mizota T, Fujiwara K. Hydration Heats of Zeolites For Evaluation of Heat Exchangers. Journal of Thermal Analysis and Calorimetry [Internet]. 2001 [cited 2022 Jul 12];64(1):157–66. Available from: <URL>.

19. Yanev Y, Cochemé JJ, Ivanova R, Grauby O, Burlet E, Pravchanska R. Zeolites and zeolitization of acid pyroclastic rocks from paroxysmal Paleogene volcanism, Eastern Rhodopes, Bulgaria. njma [Internet]. 2006 Aug 18 [cited 2022 Jul 12];182(3):265–83. Available from: <URL>.

20. Nikolov A, Rostovsky I, Nugteren H. Geopolymer materials based on natural zeolite. Case Studies in Construction Materials [Internet]. 2017 Jun [cited 2022 Jul 12];6:198–205. Available from: <URL>.

21. Ignatov I. Aluminosilicate Mineral Zeolite. Interaction of Water Molecules in Table and Mountain Zeolite Water Sevtopolis from Bulgaria. Journal of Medicine, Physiology and Biophysics. 2017;31:41–5.

22. Anonymous. Methods for evaluation of physicochemical indicators according to Ordinance No. 9 / 2001. 2004 Jul. (Official State Gazette).

23. Popova T, Petrova T, Kaleva M, Karadzhov S. Comparative study of the effect of electrochemically activated water solutions on Pseudomonas aeruginosa. Acta Microbiologica Bulgarica. 2018;34(3):160–4.

24. Popova TP, Petrova TE, Petrichev M, Valyova M. Action of activated waters on plants after adverse chemical effects, imitating acid rain. Bulgarian Journal of Agriculture Science. 2019;25(4):638–45.

25. Popova T, Ignatov I, Huether F, Petrova T. Antimicrobial activity of colloidal nanosilver 24 ppm in vitro. Bulgarian Chemical Communications. 2021;53(3):365–70.

26. Popova T, Ignatov I. In vitro Antimicrobial activity of colloidal nano silver. Bulg J Vet Med. 2021;

27. Kwakye-Awuah B, Williams C, Kenward MA, Radecka I. Antimicrobial action and efficiency of silver-loaded zeolite X. J Appl Microbiol [Internet]. 2008 May [cited 2022 Jul 12];104(5):1516–24. Available from: <URL>.

28. Matsuura T, Abe Y, Sato Y, Okamoto K, Ueshige M, Akagawa Y. Prolonged antimicrobial effect of tissue conditioners containing silver-zeolite. Journal of Dentistry [Internet]. 1997 Sep [cited 2022 Jul 12];25(5):373–7. Available from: <URL>.

29. Odabaş ME, Çinar Ç, Akça G, Araz İ, Ulusu T, Yücel H. Short-term antimicrobial properties of mineral trioxide aggregate with incorporated silver-zeolite: MTA with incorporated silver-zeolite. Dental Traumatology [Internet]. 2011 Jun [cited 2022 Jul 12];27(3):189–94. Available from: <URL>.

30. Abe Y, Ishii M, Takeuchi M, Ueshige M, Tanaka S, Akagawa Y. Effect of saliva on an antimicrobial tissue conditioner containing silver-zeolite. J Oral Rehabil [Internet]. 2004 Jun [cited 2022 Jul 12];31(6):568–73. Available from: <URL>.

31. Rivera-Garza M, Olguín MT, García-Sosa I, Alcántara D, Rodríguez-Fuentes G. Silver supported on natural Mexican zeolite as an antibacterial material. Microporous and Mesoporous Materials [Internet]. 2000 Oct [cited 2022 Jul 12];39(3):431–44. Available from: <URL>.

32. Nicholas N. How is zeolite water treatment media used in water and wastewater treatment? [Internet]. Water onine. 2019. Available from: <URL>.

33. Demirci S, Ustaoğlu Z, Yılmazer GA, Sahin F, Baç N. Antimicrobial Properties of Zeolite-X and Zeolite-A Ion-Exchanged with Silver, Copper, and Zinc Against a Broad Range of Microorganisms. Appl Biochem Biotechnol [Internet]. 2014 Feb [cited 2022 Jul 12];172(3):1652–62. Available from: <URL>.