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Standards for the Research and Registration of Drinking Mineral and Mountain Spring Waters in Bulgaria

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Author's contribution

The sole author designed, analysed, interpreted and prepared the manuscript.

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ABSTRACT

In Bulgaria is observed a great variety of spring waters. They are mineral and mountain spring waters. According to their temperature they can be cold (up to 37 °C), warm (from 37°C to 60 °C) and hot (over 60 °C). This is temperature standard in Bulgaria, European Union.
The mountain spring waters are cold with temperature up to 25 °C.
In Bulgaria for drinking mineral and mountain spring waters are valid Ordinance No 9 / 2001, Official State Gazette, issue 30, and decree No. 178 / 23.07.2004. There are include:
1. Physicochemical parameters;
2. Microbiological data;
3. Radiological study.
The aim of the publication is to show the results regarding Ordinance No. 9 / 2001, Official State Gazette, issue 30, and decree No 178 / 23.07.2004 with research of Nedyalka Valcheva, Trakia University, Stara Zagora. There are also including author research.
By their chemical composition they fall into three categories – low mineralized (up to 2 g/L), moderately mineralized (2 to 15 g/L) and highly mineralized (15 - 30 g/L). According to their chemical composition the mineral waters are divided into sulphate, sulfide, hydrogen carbonate, chloride and carbonic containing waters.

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Keywords: Mineral waters; mountain spring waters; physicochemical parameters; microbiological data; radiological parameters.

1. INTRODUCTION

In the current studies mineral waters and mountain spring waters from mountain regions of Bulgaria have been studied. It is well known that in the mountain areas of Bulgaria live the most of long-living people and centenarians. The studies are conducted by microbiological laboratory of Trakia University, Stara Zagora headed by Nedyalka Valcheva, accredited laboratory Eurotest Control, and the laboratory of Scientific Research Center of Medical Biophysics.

We analyze springs, which were examined in respect of microbiological composition and correspond 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

Analyses of the following bacteria were performed – *Total Coliforms, Escherichia coli, Sulphite reducing anaerobic bacteria (Clostridium perfringens), Pseudomonas aeruginosa*.

In physicochemical analyses are including 32-34 parameters and additionally - Hydrocarbonates (HCO_3^-), Carbonates (CO_3^{2-}), Potassium (K).

2. MATHERIALS AND METHODS

2.1 Methods for Physicochemical Analysis

1. Method for determination of color according to Rublyovska Scale – method by Bulgarian State Standard (BDS) 8451: 1977
2. Method for determination of smell at 20°C – method BDS 8451: 1977 technical device – glass mercury thermometer, conditions No 21;
3. Method for determination of turbidity – EN ISO 7027, technical device turbidimeter type TURB 355 IR ID No 200807088;
4. Method for determination of pH – BDS 3424: 1981, technical device pH meter type UB10 ID NoUB10128148;
5. Method for determination of oxidisability – BDS 3413 : 1981;
   Method for determination of chlorides – BDS 3414 : 1980;
6. Method for determination of nitrates – Validated Laboratory Method (VLM) – NO_3 – No 2, technical device photometer "NOVA 60 A" ID No 08450505;
7. Method for determination of nitrites – VLM NO_2 – No 3, technical device photometer "NOVA 60 A" ID No 08450505;
8. Method for determination of ammonium ions – VLM – NH_4 – No 1, technical device photometer "NOVA 60 A" ID No 08450505;
9. Method for determination of general hardness – BDS ISO 6058;
10. Method for determination of sulphates – VLM - SO_4 – No 4, technical device photometer "NOVA 60 A" ID No 08450505;
11. Method for determination of calcium (Ca) – BDS ISO 6058;
12. Method for determination of magnesium (Mg) – BDS 7211 : 1982;
13. Method for determination of phosphates – VLM - PO_4– No 5, technical device photometer "NOVA 60 A" ID No 08450505;
14. Method for determination of manganese (Mn) – VLM – Mn – No 7, technical device photometer "NOVA 60 A" ID № 08450505;
15. Method for determination of iron (Fe) – VLM – Fe – No 6, technical device photometer "NOVA 60 A" ID No 08450505;
16. Method for determination of fluorides – VLM – F – No 8, technical device photometer "NOVA 60 A" ID No 08450505;
17. Method for determination of electrical conductivity – BDS EN 27888, technical device – conductivity meter inoLab cond 720 ID No 11081137.

2.2 Methods for Microbiological Research

2.2.1 Nutrient media

1. Nutrientagar (MPA) with contents (in %) – meat water, peptone – 1%, agar –agar – 2% . Endo's Medium (for defining of *Escherichia coli* and *coliform* bacteria) with contents (g/dm^3) – peptone– 5,0 ; triptone– 5,0 ; lactose – 10,0 ; Na_2SO_3 – 1,4 ; K_2HPO_4– 3,0 ; fuchsine– 0,14 ; agar – agar– 12,0 pH 7,5 – 7,7 .
2. Nutrient gelatine (MPD) (for defining of *Pseudomonas aeruginosa*) with contents (in%) – Peptic digest of animal tissue; 25 % gelatin ; pH = 7, 0 – 7, 2.
3. Medium for defining of enterococci (esculin – bile agar).
4. Medium for defining of sulphite reducing bacteria (Iron Sulfite Modified Agar).

5. Wilson medium (for defining of sulphite reducing spore anaerobes (*Clostridium perfringens*) with contents(g/dm^3) – 3% Nutrient agar; 100 cm^{3}20% solution Na_2SO_3 ; 50 cm^3 20% glucose solution; 10 cm^3 8% solution ofFe_2SO_4.

2.2.2 Methods for determination of microbiological indicators

1. Methods for evaluation of microbiological indicators according to Ordinance № 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 sulphite reducing spore anaerobes – BDS EN 26461 – 2 : 2004;
5. Method for determination of 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 sulphite reducing anaerobic bacteria (*Clostridium perfringens*) – membrane method.

2.3 Methods for Radiological Study

The study including the following indicators:
Total a-activity in Bq/L
Radon in Bq/L
Total p-activity in Bq/L
Natural uranium in mg/L
Radium-226 in Bq/L
Total indicative dose in mSv/year

2.4 Standards

Ordinance No.9/2001,Official State Gazette,issue 30.
Decree No. 178 / 23.07.2004 about the quality of water, intended for drinking purposes.
BDS8451 : 1977 – defining of colour according to Rublyovska Scale, determination of smell at 20 °C.
EN ISO 7027 –determination of turbidity.
BDS3424 : 1981 – determination of pH.
BDS3413 : 1981 –determination of oxidisability.
BDS3414 : 1980 –determination of chlorides.
BDS ISO 6058 – determination of calcium, determination of general hardness.
BDS EN 27888 – determination of electrical conductivity.
VLM – NH_4 – No. 1 –determination of ammonium ions.
VLM –NO_3 – No. 2 –determination of nitrates.
VLM – NO_2 – No. 3 – determination of nitrites.
VLM– SO_4 – No. 4 –determination of sulphates.
VLM– PO_4 – No. 5 – determination of phosphates.
VLM– Fe – No. 6 – determination of iron.
VLM–Mn– No. 7 – determination of manganese.
VLM– F – No. 8 – determination of fluorides.
BDS 7211 : 1982 – determination of magnesium.
BDSEN ISO 7899 – 2 –determination of nitrates.
BDSEN ISO 9308 – 1: 2004 – determination of*Escherichia coli*andcoliformbacteria.
BDSEN26461–2:2004– determination of sulphite reducing anaerobic bacteria*(Clostridiumperfringens)*.
BDSEN ISO 16266 – determination of*Pseudomonas aeruginosa.*
BDSEN ISO 7899 – 2 – determination of eneterococci.
BDS EN ISO 6222 : 2002 – determination of total number of aerobic and facultative anaerobic bacteria.

3. RESULTS AND DISCUSSION

Physicochemical, microbiological and radiological research is conducted of mineral and mountain spring waters in Northern and Southern Bulgaria.

In Southern Bulgaria examined springs in the regions of Plovdiv [1], Haskovo [2,3,4], Stara Zagora [5,6], Sliven [7], Burgas [8], Blagoevgrad (Rupite) [9].

Performed are specific microbiological studies of springs from Southern Bulgaria [10,11,12].

In Northern Bulgaria examined mineral springs in the regions of Varna [13,14], Lovech [15] and Pleven. In Northern Bulgaria there is a great variety of mountain spring waters. The highest number of springs tested is in municipalities of Teteven [16], Yablanitsa [17] and Ugarchin [18], Lovech region.

The properties of mountain spring water are owed to its purity from snow and ice melting [19]. One of its unique features is the availability of additional energy among the hydrogen bonds in

their transition from a solid to a liquid phase [20]. Examined water with such properties is without any presence of pathogenic micro-organisms. One of the secrets of longevity is the microbiological purity of the water and the availability of the following minerals – Calcium (Ca), Magnesium (Mg), Zinc (Zn) and Manganese (Mn) [21,22].

In many areas the long-livers and centenarians consume mineral water rich in Potassium (K) and Sodium (Na) [23,24]. The six indicated minerals support the balance in the human body and metabolism, and antioxidant effects occur [25,26].

Based on the conducted physicochemical and microbiological evaluations it is established that from the four examined springs at the territory of Burgas District by physicochemical parameters thermal spring water village of Shivarovo, thermal spring water village of Polyanovo

Table 1. shows bacteria during study and microbiological parameters

Type of bacteria	Norm of measuring unit	Limit value
Escherichia coli	100	cfu/cm^3
Coliforms	100	cfu/cm^3
Clostridium perfringens	100	cfu/cm^3
Pseudomonas aeruginosa	100	cfu/cm^3

Table 2. Shows the springs by regions, which correspond to Ordinance No. 9 / 2001, Official State Gazette, issue 30, and decree No. 178 / 23.07.2004

Region	Spring
Sliven	Sliven Mineral baths; Hadji Dimitar, Shivachevo; Banya; Gunchov Spring, Karandila locality, Sliven; Nova Zagora
Varna	Drilling No P83-St.St. Konstantin and Elena; P-1x-Aquarium; P-106 x Dom Mladost; P-161xPrimorski;
Burgas	Burgas Mineral Baths; Shivarovo; Polyanovo; Drilling No B73-Medovo; Drilling No B73-Kamenar;
Yambol	Karavelovo; Stefan Karadzhovo;
Haskovo	Drilling No. 2VP, Drilling No. 3VP, KEI No. 5
Stara Zagora	Drilling No. K-3, Ovoshtnik, Drilling No. SZ-37, Yagoda; Trakia, St. Nikolay, Holly Mother of God; Center Maglij; Kazanlak; Kran-Enina; Ayazmo; Trite Chuchura; Pavel Banya - Drilling No. SZ-7; Drilling No. SZ-8; Drilling No. X-19; Drilling No. 3;
Plovdiv	Drilling No. 16-Lenovo; Drilling No. 1-Asenovgrad; Badjova voda. Hisar – Key Momina Banya; Kei Momina Salza; KEI Stublata; KEI Toplitsata; Svejest; Bistritsa; Bancheto Miromir; Choban Chesma; Chair Banya; Drilling No. 1; Staro Zhelezare Drilling No. 2; Staro Zhelezare Drilling No. 4; Drilling No. 3; Drilling No. 5; Drilling No. 6; Drilling No. 7; Parilkite; Bulgarian Rose; Narechenski bani – KEI Banski Kaptaj; Soleno izvorche. Banya – KEI Tsentralen Kaptaj; Jensko Banche; Drilling No. 1 – Kokalche; Drilling No. 8 – Dragoynovo; Drilling No. 8
Pazrdzhik	No. 2 -Vetren dol; Strelcha Velingrad – Drilling No. 5 Syarna banya; Drilling No. 3 Mizinka; Drilling No. 7 – Veliova banya; Varvara - Drilling No. 3 - Varvara; Drilling No. 5 - Varvara; Drilling No. 6 – Varvara. Banya – KEI No. 1 - Bancheto; KEI No. 2-Vetren dol; KEI No. 1 - Bancheto;
Blagoevgrad	Rupite
Sofia	Drilling No. 1- Ivanyane; Drilling No. 3- Gorna Banya; KEI Pchelinski bani; Sofia - Center
Pleven	Dolni Dabnik - Gradina
Lovech	Teteven – Dolnata cheshma; Gornata cheshma; Sondata; Klindiovo; Babintsi; Gechovoto. Golyam izvor – Tulyushovets; Krivina Troyan – Shipkovo; Chiflik. Letnitsa – Krushuna

Table 3. Shows the physicochemical parameters of mountain spring Teteven

Controlled parameter	Measuring unit	Maximum limit value	Result
1. pH	pH values	≥ 6,5 и ≤ 9,5	8.05±0.11
2. Electrical conductivity	μS/ dm^3	2000	294±9
3. Total hardness	mgekv/ dm^3	12	3.40±0.17
4. Color	Chromaticity values	Acceptable	7
5. Turbidity	FNU	Acceptable	<1.0
6. Permanent Oxidation	mgO_2/dm^3	5.0	<0.50
7. Odor	force	Acceptable	0
8. Potassium (K)	mg/ dm^3	-	0.48±0.05
9. Sodium (Na)	mg/ dm^3	200	0.72±0.07
10. Calcium (Ca)	mg/ dm^3	150	67±3
11. Magnesium (Mg)	mg/ dm^3	80	1.47±0.07
12. Zinc (Zn)	mg/ dm^3	4.0	<0.001
13. Iron (Fe)	μg/ dm^3	200	<0.1
14. Manganese (Mn)	μg/ dm^3	50	<0.1
15. Ammonium ion (NH_4^+)	mg/ dm^3	0.50	<0.013
16. Hydrocarbonates(HCO_3^-)	mg/ dm^3	-	198±10
17. Carbonates (CO_3^{2-})	mg/ dm^3	-	<12
18. Sulphates (SO_4^{2-})	mg/ dm^3	250	10.7±0.6
19. Phosphates (PO_4)	mg/ dm^3	0.5	<0.10
20. Chlorides (Cl^-)	mg/ dm^3	250	1.0±0.1
21. Fluorides (F^-)	mg/ dm^3	1.5	<0.1
22. Nitrates (NO_3^-)	mg/ dm^3	50	1.8±0.3
23. Nitrites (NO_2^-)	mg/ dm^3	0.5	<0.05
24. Mercury (Hg)	μg/ dm^3	1.0	<0.05
25. Cadmium (Cd)	μg/ dm^3	10	<0.02
26. Copper (Cu)	mg/ dm^3	2.0	<0.0003
27. Nickel (Ni)	μg/ dm^3	20	<2.0
28. Lead (Pb)	μg/ dm^3	10	<2.0
29. Aluminum (Al)	μg/ dm^3	200	<3.0
30. Antimony (Sb)	μg/ dm^3	5.0	<1.0
31. Arsenic (As)	μg/ dm^3	10	<3.0
32. Boron (B)	mg/ dm^3	1.0	<0.003
33. Selenium (Se)	μg/ dm^3	10	<3.0
34. Chromium (Cr)	μg/ dm^3	50	<1.0

Table 4. Shows the microbiological parameters of the spring sources from Burgas

Indicators	Measuring unit	Thermal spring Burgas Mineral baths with water temperature of 41°C	Thermal spring village of Shivarovo with water temperature of 41 °C	Thermal spring village of Polyanovo with water temperature of 51 °C	Thermal spring village of Judge with water temperature of 51 °C
Coliforms	cfu/cm^3	0/100	0/100	0/100	10/100
Escherichiacoli	cfu/cm^3	0/100	0/100	0/100	10/100
Enterococci	cfu/cm^3	0/100	8/100	0/100	8/100
Sulphite reducing anaerobic bacteria *(Clostridium perfringens)*	cfu/cm^3	0/100	0/100	0/100	0/100
Pseudomonas aeruginosa	cfu/cm^3	0/250	0/250	0/250	0/250

swimming pool correspond to all controlled parameters 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. With regards to microbiological parameters thermal water Burgas Mineral baths, thermal spring village of Shivarovo, thermal spring water village of Polyanovo swimming pool are in compliance with the requirements for drinking water.

4. CONCLUSION

In the present article analyses are presented of studies with regards to microbiological parameters of mineral and mountain spring waters in Bulgaria, conducted by Laboratory in microbiology, Trakia University, Bulgaria with a leader Nedyalka Valcheva. In Southern Bulgaria are examined springs in the regions of Plovdiv, Haskovo, Stara Zagora, Sliven, Burgas. Blagoevgrad.

Specific microbiological studies are performed of springs from Southern Bulgaria.

In Northern Bulgaria are examined mineral springs in the regions of Varna, Lovech and Pleven. In Northern Bulgaria there is a great variety of mountain spring waters. The highest number of springs tested is in municipalities of Teteven, Yablanitsa and Ugarchin, Lovech region.

The purity of stated springs complies with 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, and Decree No. 14 regarding the resort resources, resort localities and resorts.

The waters that correspond to Ordinance No. 9 / 2001 are suitable for drinking consumption.

COMPETING INTERESTS

Author has declared that no competing interests exist.

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