

Water Quality of Public Wells from Rural Area of Ampoi Basin, Transylvania

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Abstract: The problem of drinking water quality, including the wells from the Ampoi basin must fulfill the European norms, especially when these waters are used by the population and animals. The aim of this paper is to assess the microbiological (number of aerobic bacteria, *Escherichia coli* and *Enterococci*) and physical-chemical parameters of 7 groundwater samples (from wells) and also time tracking of changes in these parameters in different seasons. The results evidence that anthropogenic pollution affect water quality from wells in the Ampoi basin. The major contamination was found in the villages Metes, Taut, Sard and Miesti. Also, for village Metes the evolution in time of these parameters was monitoring.

Keywords: groundwater, wells, quality, rural area

1. Introduction

Water in nature is never pure; given the interactions with the environment, it contains gases, mineral and organic substances, dissolved in suspension. Drinking water must be health providing, clean, without microorganisms, parasites or substances which, by number or concentration, can be a potential hazard for human and animal health [1].

Underground waters are an important resource, taking into account that unlike surface waters they are usually less polluted or even unpolluted. Therefore, underground waters may be made potable using minimal measures, sometimes using only disinfection, or without any treatment [2].

In rural areas that don't have distribution of potable water through public supply systems, the wells that represent sources of groundwater are very important. This water is not treated and is often subjected to chemical and microbiological pollution from anthropogenic sources [3].

The ground water in rural areas can be polluted as a result of farming activities. The quality of the drinking water drawn in wells can be affected especially for those wells that are placed in the valleys, and at insufficient distance from the stables [4-5]. Water quality assessment is made by measuring of certain parameters (physical, chemical and microbiological) whose limits are legally defined [6]. The drinking water quality, is set by Law no. 311/2004 who complete the Law no. 458/2002 (in Romania), these laws transposing the Directive 98/83/EC.

Biological pollution of water is made through the sewage from hospitals, human and veterinary clinics, diagnostic laboratories, slaughterhouses, rendering plants [7]. The polluted water is a source of infections and parasitic infestations not only for animals but for people too, and the bacterial pathogens that are transmitted

through water to humans and animals cause bacterioses.

The present paper seeks the ways of testing the quality of drinking water, especially wells water. The study results show that the organoleptic, physico-chemical and microbiological parameters of wells water (public sources) from the Ampoi basin have values that exceed the limits for potable drinking water, only with some minor exceptions.

2. Experimental

In figure 1, the map area of Ampoi basin and the sampling points of public wells water are presented.



Figure 1. The map of sampling points

For the study, seven water samples were collected in November 2012 from public wells located in the following villages from Alba County: Fenes, Presaca Ampoiului, Poiana Ampoiului, Metes, Taut, Sard and Miesti, all situated in Ampoi basin. For village Metes sample, the evolution of the parameters over time was monitoring.

The pH values were measured with the InoLab pH-meter. The amount of total hardness present in the given

sample of water was estimate by EDTA titration method [8]. The acidity and alkalinity (AT) was determinate using method ASTM D1067 – 11 [9].

Determination of total number of bacteria growing at 37°C, 22°C (mesophyll) [10-11]: the method consists in inoculation, by including, of a quantity of 1-2 mL from the sample or decimal dilutions (10^{-1} and 10^{-2}), into a Petri plate in 10-15 cm³ nutritive gellose (melted and cooled at 45°C); after the solidification of the gellose the plates are incubated 37±2°C, for 44±4 h. The colonies are counted both those at the surface, and the ones within the gellose.

Detection and counting of Escherichia coli and coliform bacteria Part 1: Membrane filtration method [12]: 100 mL from the water sample are filteres through a membrane settled on lactose TTC agar: all was incubated at 36±2°C, 21±3 h. It was counted as lactoso-positive bacteria the colonies developing on yellow medium. The confirmation: a) Oxidase test: 10 characteristic colonies are passed on nonselective agar (TSA) and incubate at 36±2°C, 21±3h; then, from each Petri plates it was take one colony and put it on filtering paper impregnated with oxidase reagent. Positive reaction: the occurence of purple blue coloration in 30 sec. b) Indole test: were made simultaneous inseminations in tryptophane broth and incubated at 44°C, 21±3h, then was added 0.2-0.3 mL of Kovac's reagent. Positive reaction: the occurrence of a purple red ring. The colonies with negative reaction to oxidase were counted as being coliform bacteria. The colonies with negative reaction to the oxidase and positive reaction with the indol were counted as being E. coli.

Identification and counting of intestinal enterococci Part 2 Membrane filtration method [13-14]: 100 mL or 10 mL from the sample and/or dilutions were filtered through the membrane placed on Slanetz Bartley medium and incubated at 36±2°C for 44±4h. The typical colonies are pink or brown, in the middle or totally. They were transferred on bile-esculin-azide agar, pre-heated at 44°C and incubated at 44°C, for 2h. The characteristic colonies which are bronze-black were counted and the calculus method from SR EN ISO 8199: 2008 was applied.

The methods of rapid spectrophotometric determinations involved the use of the spectrophotometer Spectroquant NOVA 60 (SQ) and SQ specific kits (with reagents and reaction tubes). Following the work pattern from the kit, the SQ was read [15].

Ammonium: Kit SQ domain 0.010-2 mg/L NH₄-N or 0.01-2.58 mg/L. 0.5 mL from the sample was dropped in the reaction tube and homogenised. A dose of NH₄-1K was added, closed, shake, and, after 15 min. read. In high alkaline solutions, the nitrogen ammonia is present almost totally as ammonia, reacts with hypochlorite ions resulting in monochloramine, which reacts with substitute fenol and forms a blue indocarbolitic derivative.

Nitrates: Kit SQ domain 1.0-50.0 mg/dm³ NO₃-N or 2.2 – 79.7 mg/dm³ NO₃⁻. 0.5 mL of test was putted in the tube. 1 mL NO₃-1K was added, shacked for a minute and let aside for 10 minutes. In sulphuric acid solution, nitrate ions react with a derivative of benzoic acid to form a red nitro compound.

Nitrites: Kit SQ 0.02-1.00 mg/dm³ NO₂-N or 0.07-3.28 NO₂⁻ NO₂ 10 mm tube. 5 mL of sample were drop in the tube. A micro palette knife of NO₂ -1 reagent was added and shacked until total dissolution. Reaction time: 10 minutes. In acid solution, nitrite ions react with the sulphanic acid resulting diazonium compound; which then reacts with N-1-naftiletilendiamine dihydro-chloride resulting a violet red nitro compound.

3. Results and Discussion

The results of organoleptic test, made according to method EN 1622/2007, have been showed that the samples can be characterized as a clear liquid, colorless, without particles and without sediment impurities. The samples didn't have foreign taste or smell, except samples taken from villages Metes, Taut and Sard, which were liquids slightly opalescent, colorless with impurities and sediment suspension, without foreign taste or odor. The sample collected from Taut has showed a slight yellowish colour.

The results of laboratory tests: physical, chemical and microbiological parameters of samples collected from public wells of Ampoi basin are presented in tables 1 and 2.

TABLE 1. Physico-chemical parameters of wells water

Parameter Source	pH	Hardness, [°dH]			AT, [mg/L]	Acidity, [mg/L]
		Temporary	Permanent	Total		
Fenes	6.9	15.1	2.1	17.2	5.4	1
Presaca Ampoiului	7.25	9.2	5.1	14.3	3.3	4
Poiana Ampoiului	7.4	2.8	24.5	27.3	1	3.5
Metes	7.7	8.4	30.8	39.2	3	7
Taut	7.2	5.6	59.4	65	2	5.5
Sard	7.5	8.9	14.2	23.1	3.2	6
Micesti	7.5	14	12.4	26.4	5	3.5

All samples had pH values between 6.5 and 9.5, representing normal values for drinking water (≥ 6.5 ; ≤ 9.5). The lowest hardness (soft water) was found in samples from Presaca Ampoiului.

Samples of public wells water from villages Presaca Ampoiului and Poiana Ampoiului were negative for the presence of microorganisms. Coliforms and E. coli were identified only in well water collected from Metes. Enterococci were identified in wells water collected from Fenes, Metes, Taut and Sard. The total number of germs that grow at 22 °C and 37 °C were above the limits stipulated by the Romanian Law 458/2002 in the samples taken from Metes, Taut, Sard and Micesti.

The figures 2, 3, 4, 5, 6, 7 and 8 show the variation in time (quarterly for the period 2010 – 2012) of physico-chemical and microbiological parameters of water samples collected from Metes public well water.

TABLE 2. Microbiological parameters of water wells

Parameter Source	UFC/mL		Total coliforms /100 mL	E.coli/ 100 mL	Enterococcus /100 mL
	22°C	37°C			
Fenes	Abs	8	Abs	Abs	2
Presaca Ampoiului	25	6	Abs	Abs	Abs
Poiana Ampoiului	22	Abs	Abs	Abs	Abs
Metes	2,0x10 ²	1,7x10 ²	120	94	9
Taut	1,2x10 ²	17	Abs	Abs	16
Sard	1,2x10 ²	18	Abs	Abs	4
Micesti	1,1 x10 ²	4	Abs	Abs	Abs
Normal values according to Law 458/2002 for bottled water	100	20	0	0	0
Normal values for mineral water at source, according to Law 1020/2005	20	5	0	0	0

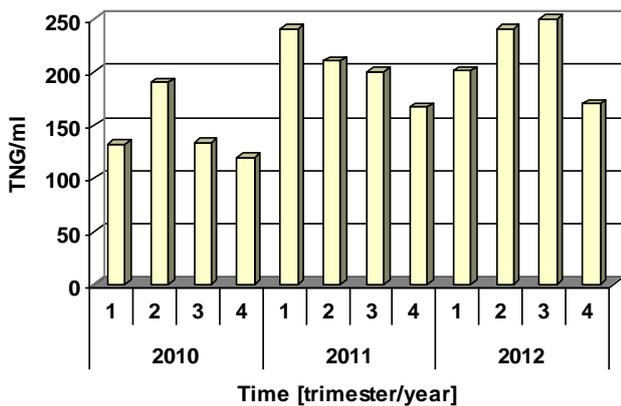


Figure 2. Time variation of total mesophilic aerobic bacteria (total number of germs) in Metes samples

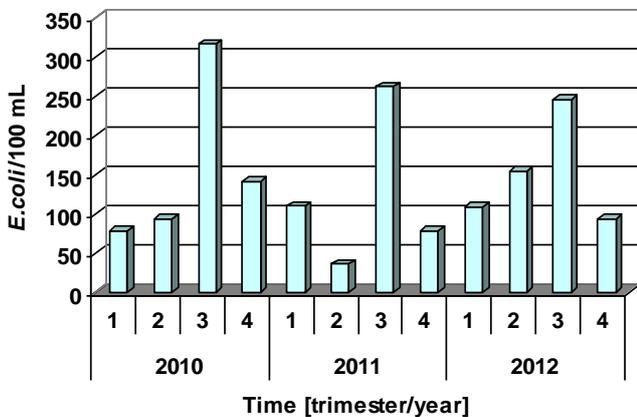


Figure 3. Time variation of E. coli in Metes samples

From figure 2 it was noticed a considerable increase of mesophilic aerobic bacteria in all periods of year, with the highest value of 250 colonies recorded in the third quarter of the year 2012. All values were higher than the limit of 100 colonies allowed by law for bottled drinking water (Directive 98/83/EC). Overall, there is a growing evolution for TNG in time (2010-2012).

All samples were positive for the presence of the micro-organism - Escherichia coli. The pollution may be caused by rain or snow that infiltrated into the ground. This indicates a contamination source due to poor planning of the well.

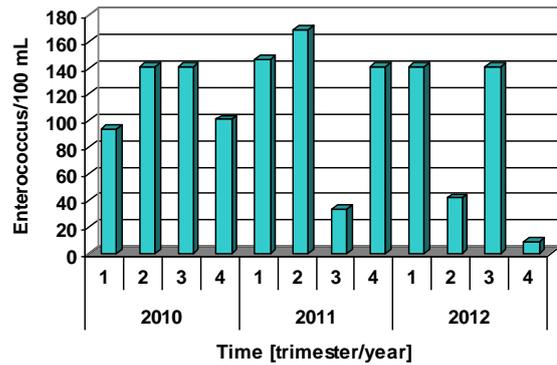


Figure 4. Time variation of enterococci in Metes samples

Enterococci were identified in wells water sampling during the three years of monitoring; the values were above the limits stipulated in Romanian Law no. 458/2002 that transposes the Directive 98/83/EC.

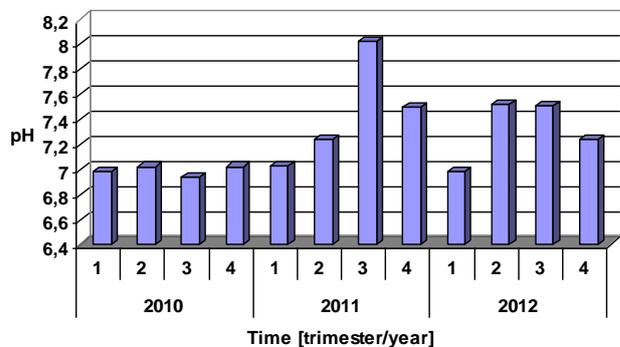


Figure 5. Time variation of pH in Metes samples

The pH values were within the limits for drinking water ($\geq 6.5, \leq 9.5$), ranging between 6.94 and 7.52.

Nitrogen in the water was present in several different forms such as molecular nitrogen, oxides of nitrogen, ammonia, ammonium compounds, nitrates and nitrites. The nitrogen, as a part of the biogeochemical cycle it is a concept that recognizes the dynamism of multiple, complex processes determined by different factors in aquatic ecosystems. Algae can use both free nitrogen from water but also salts of ammonia (NH_3) and after their exhaustion, nitrates (NO_3^-). Bacteria play an important role for nitrogen cycle in aquatic ecosystems and in the bacterial transformation of nitrogen compounds which are reversible. All processes are dependent on the amount of oxygen dissolved in water.

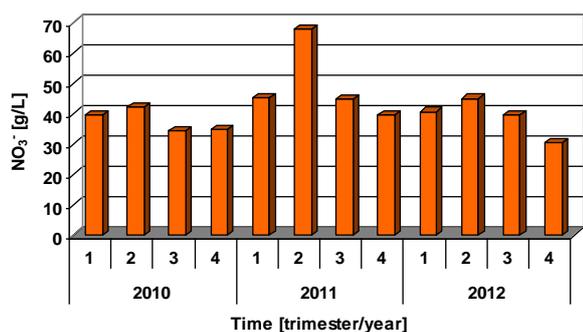


Figure 6. Time variation of nitrates in Metes samples

The NO_3^- [mg/L] content of water doesn't exceed the accepted limit of 50 mg/L for drinking waters. The exception was the sample collected in the second quarter of 2011.

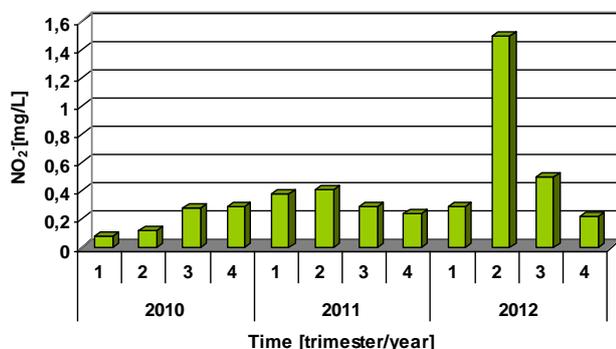


Figure 7. Time variation of nitrates content in Metes samples

The samples collected from public well situated in village Metes did not exceed the content of nitrites (NO_2^- [mg/L]), except the sampled analyzed in second and third quarter of year 2012. According to Directive 98/83/EC, the maximum value for drinking waters is 0.50 mg/L. Nitrites are found due to the pollution of water with organic matter, or by partial oxidation of the amino radical or by reducing nitrate. Their presence indicates an older pollution of water, but together with high concentrations of ammonia they point out that the pollution is continuous.

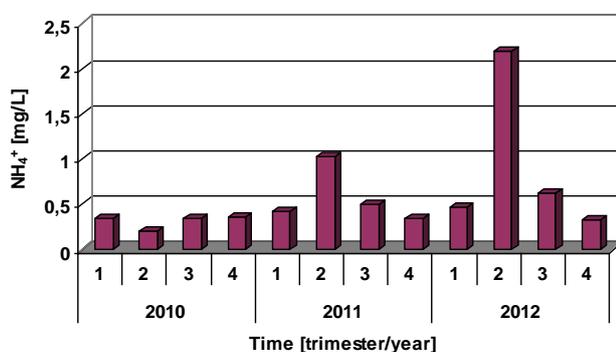


Figure 8. Time variation of ammonium content in Metes samples

The values of ammonium ions are below the law limit, except the sample from the second quarter of 2011 and second and third quarter of 2012, when it exceeded the maximum permissible value of 0.50 mg/L. Increases are recorded in warm season and the trend is the increase in time of values. Ammonia occurs after water pollution by organic substances that undergo decomposition, being the first step of nitrogenous substances degradation. Its presence indicates a recent pollution.

Statistical analysis

The current trend of research is characterized by using of computer in the study of different phenomenon related to the chemistry, biochemistry, physics, etc. Practical use involves the processing of experimental data for simulation of chemical or biological phenomena. In chemistry is working with models of various types. In this study a model $y=f(x_1, x_2)$ was used. The main parameter (dependent variable) (y) was considered UFC/mL. The secondary parameters (independent variables) were considered: x_1 =time [months], x_2 =a) NH_4^+ content [mg/L], b) NO_3^- content [mg/L], c) NO_2^- content [mg/L].

Experimental measurements revealed a series of data that can be used to evaluate the comparative effect of measured parameters by performing a multiple correlation analysis of microbial growth as the dependent variable, depending on other parameters: ammonium ion content, nitrates and nitrites in time, were the last parameters are independent variables.

Nitrogen, nutrient of great importance in aquatic ecosystems, enters in water by several ways and is found in water in many forms: molecular nitrogen, nitrogen oxides, ammonia, ammonium, nitrites and nitrates. So nitrogen biochemistry involves taking into account all its oxidation states, from 5 to 0 and -3. [16]

In ecosystem, nitrogen enters in the biogeochemical cycle, determined by a complex of interactional factors from aquatic ecosystems. Algae can use the free nitrogen from water and ammonia salts (NH_3) and after their exhaustion even the nitrate (NO_3^-) [17].

By using STATISTICA 6.0 software, experimental data were processed and analyzed, generating statistical models that establish the connection between the microorganisms presence and other parameters as: ammonium ion content, nitrates and nitrites content in time.

For the samples taken from the Metes village, the equations presented in table 3 we obtained:

TABLE 3. Equations of statistical mathematical models

Case	Equations of statistical mathematical models
a)	$y = 43.95 + 7.04 \cdot x_1 + 2.39 \cdot x_2$
b)	$y = 141.08 + 4.89 \cdot x_1 + 45.99 \cdot x_2$
c)	$y = 138.66 + 4.89 \cdot x_1 + 29.28 \cdot x_2$

The equations are valid on the studied values range. After calculating the model coefficients, it is necessary to make a comparison between model predictions and experimental data (table 4).

TABLE 4. Indicators of statistical models adequacy

Case	Variance, σ^2	Indicator of the model precision, R^2	Correlation coefficient, R
a)	33.44	0.53	0.73
b)	37.73	0.40	0.64
c)	37.91	0.40	0.63

In the case of first-order polynomial equation, the indicators of model adequacy show a satisfactory correlation between the considered variables.

4. Conclusions

The results obtained evidence that anthropogenic pollution affect water quality from wells in the Ampoi basin. There is a major water contamination especially in the villages Metes, Taut, Sard and Micesti. It is clear that all activities taking place on the surface have an impact on groundwater quality.

The study shows that the risk due to consumption of water from wells in the Ampoi basin is reduced, but not completely. There are water sources that exceed the limits allowed by legislation for microbiological parameters - in particular.

An important role in ensuring water quality in rural areas is the introduction of centralized water supply system and of sewerages implicitly to avoid groundwater contamination.

The equations of statistical models can approximate microbiological growth in wells water knowing the time of the year when the sample was collected and its content of ammonium ion, nitrates or nitrites. Correlation parameters calculated based each on other arguments a satisfactory capacity of prediction for the mathematical statistical models. Also, the model predictions can constitute a control criterion for assessing groundwater quality.

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