

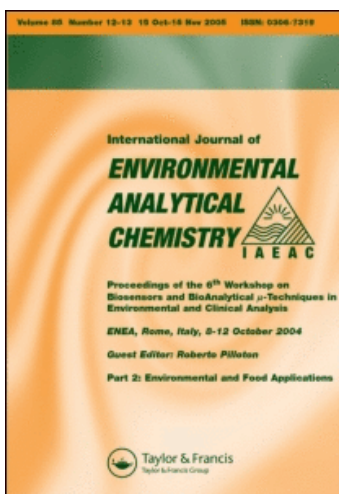
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The quality of drinking water supplies in North-Western Greece: a three-year follow-up

Nikolaos Giannoulis^a, Vasiliki Maipa^a, Triantafyllos Albanis^b, Ioannis Konstantinou^b, Ioannis Dimoliatis^a

^a Department of Hygiene and Epidemiology, Medical School, University of Ioannina, 45110 Ioannina, Greece ^b Department of Chemistry, Laboratory of Environmental Technology, University of Ioannina, 45110 Ioannina, Greece

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THE QUALITY OF DRINKING WATER SUPPLIES IN NORTH-WESTERN GREECE: A THREE-YEAR FOLLOW-UP

NIKOLAOS GIANNOULIS^a, VASILIKI MAIPA^a,
TRIANAFYLLOS ALBANIS^b, IOANNIS KONSTANTINOU^b
and IOANNIS DIMOLIATIS^{a,*}

^a*Department of Hygiene and Epidemiology, Medical School,
University of Ioannina, 45110 Ioannina, Greece;* ^b*Department of Chemistry,
Laboratory of Environmental Technology, University of Ioannina, 45110 Ioannina, Greece*

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The study was undertaken to assess the microbiological and physicochemical quality of potable water of Arta, Preveza and Lefkada prefectures in North-Western Greece, during a 36-month survey (1996–1999). Drinking-water samples were collected from twelve points along the distribution networks located at the three cities of Arta, Preveza and Lefkada. The drinking-water quality standards were analyzed with respect to the presence of *total coliforms* (TC), *fecal (thermotolerant) coliforms* (FC) and *fecal streptococci* (FS). Some physicochemical parameters such as temperature, pH and total dissolved solids (TDS) were also determined. Standard techniques for water sample collection and analysis set by the American Public Health Association were used. Microbiological analyses indicate that of the 456 samples analyzed along the distribution network from the springs to the consumer potable tap, the individual failure rates were 35.1, 27.4 and 12.3% for TC, FC and FS, respectively. The combined failure rate according to the limit set by the 80/778 directive of the European Union was 37.9% for the analyzed samples. Failure rates on microbiological indicators displayed a seasonal trend being greater during the autumn–winter period. Although this observation is likely due to a combination of local and regional scale factors, a part of the variability in the failure rate was explained by a significant positive relationship with the rainfall amount. The results showed that there are considerable variations among the examined samples with respect to their physicochemical properties, which lie below the maximum permissible levels of the European drinking water standards. A higher failure rate for the samples collected directly from the springs compared with those taken from the potable tap suggests that the groundwater itself contributes much of the microbiological contamination and physicochemical alterations rather than the storage or a supply line contamination mechanism.

Keywords: Drinking water; Groundwater; Contamination; Coliforms; Streptococci; Greece

INTRODUCTION

The provision of adequate and safe drinking water is a high-priority issue and a basic necessity for safeguarding the health and well-being of humans all over the world. The World Health Organization (WHO) reported that nearly half of the population in

*Corresponding author. Fax: +32-6510-97853. E-mail: idimolia@cc.uoi.gr

developing countries suffers from health problems associated with lack of drinking water or with microbiologically contaminated water [1]. Traditionally, the microbiological quality of drinking water has been the main concern. Groundwater represents an important source of drinking water and its quality is currently threatened by the combination of over-abstraction, chemical pollution and microbiological contamination [2]. There are many sources of groundwater contamination, but broadly they can be divided into two categories: point source and diffuse source (treatment and distribution) contamination [3, 4]. The regional scale composition of groundwater has been affected by human activities through changes in land use and intervention in natural flow patterns [5]. The available data about the quality of European groundwater sources are largely variable and scrappy owing to the lack of either a harmonized monitoring strategy or an information network [6]. In Greece the majority of the urban population receive potable water from a variety of sources, including groundwater, through the regional authorities. The nature of the water sources, the method of supply and the potential risk from contamination vary widely. The information describing the microbiological quality of drinking water is increasing but remains insufficient to adequately distinguish the natural variability in groundwater quality and therefore the adequacy of sampling frequencies [7].

The potential health concerns associated with contaminated water supplies can be broadly grouped into those associated with contamination by microbiological agents and/or those associated with chemical agents. In areas dominated by agricultural activities there is the additional risk of diffuse pollution by agrochemicals such as nitrate and pesticides [8, 9].

As far as distribution networks are concerned, drinking water contains organic and inorganic matter, which can accumulate to the surface of drinking-water pipelines. Microbes can easily penetrate into the drinking-water distribution network, utilizing these substrates [10]. Authorities responsible for water intakes and distribution facilities should carry out routine monitoring of drinking-water quality, but the water quality of wells and springs in rural areas is only randomly checked.

Coliform bacteria, inhabitants of the intestinal tract of warm-blooded animals, have traditionally been used as indicators of water quality. It is assumed that the possibility of the presence of pathogens increases with pollution by feces and urine. The most frequently used method for the determination of fecal pollution of a water sample is to test for the presence of fecal indicator bacteria. Other than coliforms, the most useful indicator organisms for fecal contamination are strains of *Streptococcus fecalis*. The total coliform group and fecal streptococci are considered excellent indicators of water bacterial contamination, because those bacteria are always present in the normal intestinal tract of humans and other warm-blooded animals and are eliminated in large numbers in fecal wastes [11–13]. Several studies have been reported on the microbiological contamination of water supplies and most of them reveal that the majority of sources fail to meet drinking-water quality standards [2, 14–18].

Drinking-water and groundwater contamination is a major public health and environmental concern in North-Western Greece because the majority of urban and rural populations use groundwater resources for drinking water. The present study focuses on the assessment of the microbiological quality of water supplies in three major towns, Arta, Preveza and Lefkada, the capitals of the corresponding prefectures, in North-Western Greece, in accordance with the 80/778 EU directive for drinking water. The values of the microbiological indicators are also correlated with the measured

physicochemical properties. The water quality was monitored through the entire distribution system, from the springs to the consumer taps, during the period 1996–1999. Samples were grouped using attributes such as sampling point and sample date in order to assess trends and relationships. Regional and seasonal patterns as well as the possible factors affecting the water quality are also discussed.

MATERIALS AND METHODS

Study Sites and Sample Collection

Figure 1 shows the region under study and the sampling points along the distribution network of water supplies. The springs of Agios Georgios (sampling station S1) flow from the carstic aquifer that is developed along the west bank of river Louros and springs off at 113 m altitude. The shallow aquifer consists of limestone formations and the mean outlet of the springs is about $2.6 \text{ m}^3/\text{s}$. Average seasonal precipitation for the region under study was 133.9, 38.1, 4.3 and 101 mm for winter, spring, summer and autumn respectively. Average precipitation for the period 1996–1999 was 277.2 mm and 85% of the precipitation occurs from September to February. The water quantity is enough to supply the population of the three town agglomerations of Arta, Preveza and Lefkada (about 100 000 inhabitants). The community of Agios Georgios, whose waste is treated in septic tanks, is situated near the springs (300 m). In addition, the village cemetery is located close to the springs. In the surrounding

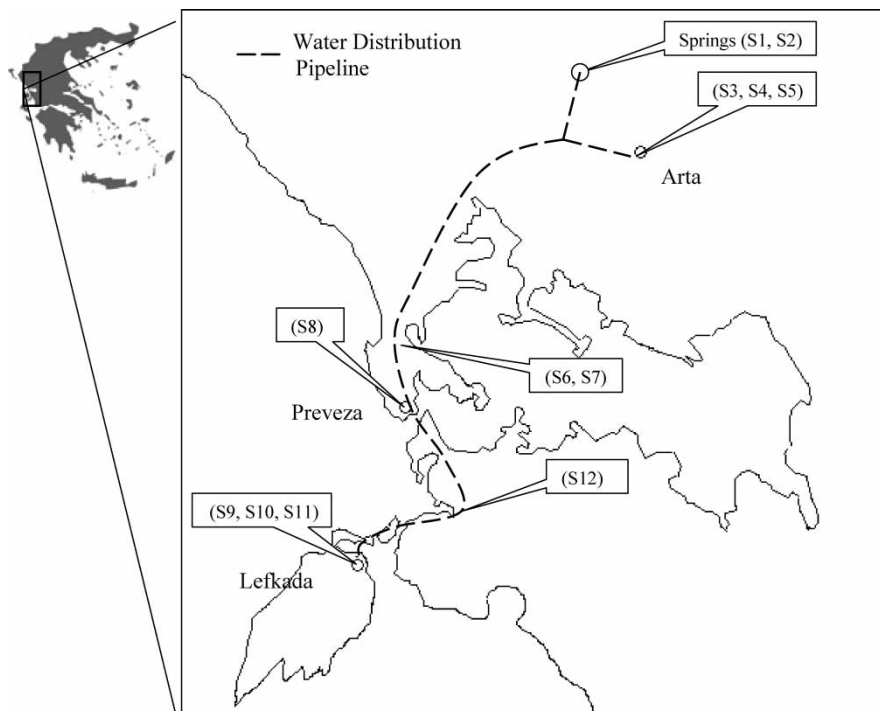


FIGURE 1 Map indicating the distribution of water supplies and the sampling stations within Arta, Preveza and Lefkada municipalities, NW Greece.

region, free and industrialized cattle activities were developed. The main animal husbandry includes pigs and sheep.

Two distribution networks are used for the distribution of water in the town agglomerations. One supplies the municipalities of Preveza and Arta and the second supplies the island of Lefkada. Before distribution, the water is disinfected with chlorine (residual chlorine range: 0.1–0.2 mg/L) and stored in concrete reservoirs. After treatment with chlorine, municipal water is pumped directly to individual homes through a piped network and stored in concrete reservoirs after treatment with chlorine. External pipelines are made of cast iron or polyethylene (PEH) and internal distribution pipelines are made of PVC and, to a small extent, asbestos cement. The sampling stations along the distribution from the springs to the end of pipeline were as follows (Fig. 1): S1 = springs; S2 = Agios Georgios, after primary disinfection; S3 = Arta, prior to disinfection; S4 = Arta, after disinfection; S5 = Arta tap, S6 = Preveza prior to disinfection; S7 = Preveza after disinfection; S8 = Preveza tap, (S9, S12) = Lefkada, prior to disinfection, S10 = Lefkada, after disinfection; S11 = Lefkada tap.

During the period 1996–1999 a total of 456 water samples for microbiological and 266 samples for physicochemical analysis were collected from different points consisting of: 76 and 38 water samples from the springs, 114 and 76 from each of the Arta and Preveza piped networks and 152 and 76 from the Lefkada piped network. The samples were taken before and after disinfection with chlorine and at the end of pipelines (consumer's tap) in order to investigate possible deterioration of the water quality originating from the distribution network itself.

Water samples were obtained in accordance with the Standard Methods for the examination of Water and Wastewater [19], and the recommendations of the World Health Organization [20]. Water samples were collected in sterile dark glass bottles and a small amount of $\text{Na}_2\text{SO}_3 \cdot 5\text{H}_2\text{O}$ solution (18 mg/L) was added in order to block the continuous disinfectant action of chlorine. The samples were kept in a portable refrigerator and were taken to the laboratory (ambient temperature 25°C) for analysis. The time between the sample collection and processing did not exceed four hours.

Physicochemical Analyses

The determinations were carried out according to the Standard Methods for the examination of Water and Wastewater [19, 21]. pH and temperature measurements were performed in the field using a portable pH-meter (Model LF 325, WTW, Weilheim, Germany) calibrated against two standard buffer solutions of known pH values (pH 3 and 10) equipped with a calibrated thermometer. The determination of total dissolved solids (TDS) was used as a general indicator for poor water quality and was performed with a conductivity meter (WTW, Germany) that is not influenced by temperature alterations.

Microbiological Analyses

All analyses were performed using the membrane filtration (MF) culture method in accordance with the Standard Methods [22]. The MF method is fully accepted and approved as a procedure for monitoring drinking water microbial quality. Each water sample was analyzed for total coliforms (TC), fecal coliforms (FC) and fecal streptococci (FS). TC bacteria were enumerated (results expressed as colony forming units

(cfu) per 100 mL) using m-Les Endo Agar (Difco Laboratories, Detroit, MI) incubated at 36°C for 24 h. Confirmation was made by selection and culturing of characteristic colonies in BGLB (Brilliant Green Lactose Broth) at 36°C for 24 h.

Fecal coliforms were enumerated using m-FC Agar (Difco Laboratories, Detroit, MI) incubated for 24 h at 44°C. Confirmation was made by selection and culturing of characteristic colonies in LTLSB (Lactose-Tryptone-Lauzyl-Sulphate-Broth) at 44°C for 24 h. Fecal streptococci were measured with the MF method on to Slanetz and Bartley Agar (Oxoid Ltd.) incubated at 36°C for 48 h. Confirmation was performed using the Escoulin hydrolysis method. The membranes were transferred on petri dishes with Esculin bile Agar (Merck) incubated at 44°C for 1 h.

Failure Rates and Statistical Analyses

In all cases samples which failed were those whose values of TC, FC or FS were >0 and/or were above the statutory limit for pH or TDS as given by the EU 80/778 Directive regulations. The results are expressed as failing samples as a percentage of the total number of samples (failure rate) analyzed. The microbiological and physicochemical data were analyzed by Spearman rank correlation analysis using the Minitab computer software (Version 13.1). Physicochemical data were presented as the arithmetic means while the numbers of microbes have been presented as median concentrations because they followed a non-normal distribution.

RESULTS AND DISCUSSION

Water Quality Status

Summary results (median, max.–min. values) of the three bacterial indicators together with the physicochemical parameters (mean, max.–min. values) in the different sampling stations are given in Table I. In all the sampling stations TC and FC were detected with a maximum value of 43 cfu/100 mL for S1 (springs). Only in two sampling stations (S8 and S11) were FS not detected and of the rest the maximum value (9 cfu/100 mL) was found again in S1. The median value of coliforms for the end-of-pipe sampling stations, of Arta, Preveza and Lefkada (S5, S8, S11) was 0 cfu/100 mL, in accordance with the limit set by the EU. Microbiological examination of the springs water samples showed a relatively high coliform count that may be attributed to contamination from human and cattle activities in their vicinity. All the physicochemical parameters studied lie within the permissible range of the current drinking-water quality standards (80/778 EU Directive).

The data presented in Table II include a summary of 456 individual samples indicating the failure rate for all the sampling stations (S1–S12) reflecting the general situation of water supplies in the three cities. The microbiological analyses of the samples along the distribution network from the springs to the consumer's potable tap indicate that the individual failure rate was 35.1, 27.4 and 12.3% for TC, FC and FS, respectively. The combined failure rate for these samples was about 37.9%. Moreover Table II shows the combined and partial failing numbers as a proportion of the samples collected for each sampling point during the period 1996–1999 according to the three bacterial indicators. TC is the dominant indicator that causes failure and FC, FS follow with lower failure

TABLE I Microbiological and physicochemical characteristics of the water samples in the period 1996–1999 ($N=266$)

	<i>S1</i>		<i>S3</i>		<i>S5</i>		<i>S6</i>		<i>S8</i>		<i>S9</i>		<i>S11</i>	
	Mean	Range	Mean	Range	Mean	Range	Mean	Range	Mean	Range	Mean	Range	Mean	Range
Physicochemical														
<i>T</i> (°C)	18.5	15.0–23.3	19.2	14.7–24.1	19.6	13.6–25.2	19.3	13.4–24.8	20.1	13.6–26.2	20.1	13.8–27.1	20.5	13.1–27.3
pH	7.1	6.7–7.4	7.2	6.9–7.5	7.3	7.1–7.5	7.3	7.1–7.5	7.3	7.1–7.5	7.3	7.1–7.5	7.3	7.2–7.4
TDS	457.8	437.6–478.4	435.2.4	457.4–479.2	460.4	434.4–488	457.8	424.8–487.2	460.3	428–483.2	462.4	435.2–480.8	463.7	435.2–484
Microbiological ^a	Median	Range	Median	Range	Median	Range	Median	Range	Median	Range	Median	Range	Median	Range
TC	4.5	0.0–43.0	0.0	0.0–42.0	0.0	0.0–18.0	1.0	0.0–38.0	0.0	0.0–21.0	2.0	0.0–25.0	0.0	0.0–28.0
FC	1.0	0.0–43.0	0.0	0.0–37.0	0.0	0.0–2.0	0.0	0.0–29.0	0.0	0.0–1.0	1.0	0.0–18.0	0.0	0.0–6.0
FS	0.0	0.0–9.0	0.0	0.0–2.0	0.0	0.0–1.0	0.0	0.0–5.0	0.0	0.0	0.0	0.0–2.0	0.0	0.0

S1 = spring, *S3* = Arta, prior to disinfection with chlorine, *S5* = Arta tap, *S6* = Preveza, prior to disinfection with chlorine, *S8* = Preveza tap, *S9* = Lefkada, prior to disinfection with chlorine, *S11* = Lefkada tap. ^aResults expressed in cfu per 100 mL.

TABLE II Microbiological quality data for Arta, Preveza and Lefkada municipal water supplies during the period 1996–1999

	Sampling point												Total
	S1	S2	S3	S4	S5	S6	S7	S8	S9	S10	S11	S12	
No. of samples collected	38	38	38	38	38	38	38	38	38	38	38	38	456
TC failure (%)	78.9	50.0	34.2	10.5	10.5	52.6	13.2	5.3	68.4	10.5	28.9	57.9	35.1
FC failure (%)	63.2	50.0	26.3	2.6	5.3	39.5	0.0	2.6	55.3	5.3	21.1	57.9	27.4
FS failure (%)	31.6	31.6	10.5	2.6	2.6	31.6	0.0	0.0	10.5	0.0	0.0	26.3	12.3
Combined failure (%)	81.6	52.6	34.2	10.5	10.5	63.2	13.2	5.3	71.1	10.5	28.9	73.7	37.9

Failures (by indicator) are shown as a percentage of the number of samples collected at each point or of the total number of samples collected.

rates. More elevated concentrations were found in the springs (S1) and before entering the distribution network of the three cities, in sampling stations S3, S6 and S9 (before disinfection with chlorine). Among the piped network samples, S11 (Lefkada) was found the most contaminated. Thus, the sample stations S1, S3, S5, S6, S8, S9 and S11 were selected as more important and indicative for the water quality monitoring.

Origins of Contamination and Seasonal Patterns of Groundwater Quality

The seasonal variation of physicochemical parameters is shown in Figs. 2 and 3. TDS concentration increases from the winter to the autumn period while the pH range is almost stable during the year. The water temperature ranged from 17.5 to 18.6°C in winter, from 17.5 to 20.6°C in spring, from 21.9 to 25.2°C during summer and from 18.8 to 20.4°C in autumn. To establish the relationship between the microbiological parameters tested in the seven sampling stations, the Spearman coefficient test was applied (Table III). In general, higher correlations among the three bacterial indicators were found during the winter and autumn period and lower for the spring and summer period. TC was correlated with FC in all seasons but TC with FS and FC with FS correlations were found only during winter and autumn. For monitoring drinking-water quality it is important to study not only the relationship between the microbiological indicators but also the influence of the environmental factors on the microbiological population (Table IV).

None of the indicator microorganisms used showed significant correlation with the physicochemical parameters during the spring and summer periods. On the contrary during autumn all the microbiological indicators were correlated with the physicochemical properties with significant although low correlations. In addition fewer but similar correlations were obtained during winter between TC and temperature and pH and between FC, FS and temperature. Low water temperature in drinking water favors the survival of microorganisms and an increase in temperature results in a rise of their activity and a reduction in their generation time [23]. This could explain the positive correlation found between the TC, FC indicators and temperature during winter and the negative correlation found during autumn. Significant positive relationships were established between the three indicators and the average seasonal rainfall as follows: $r = 0.622$, $r = 0.581$, $r = 0.594$ for TC, FC and FS respectively, with $p < 0.001$.

Despite the similar number of monthly samples analyzed, a marked seasonality in the proportion of samples failing for microbiological parameters was observed (Figs. 4–6).

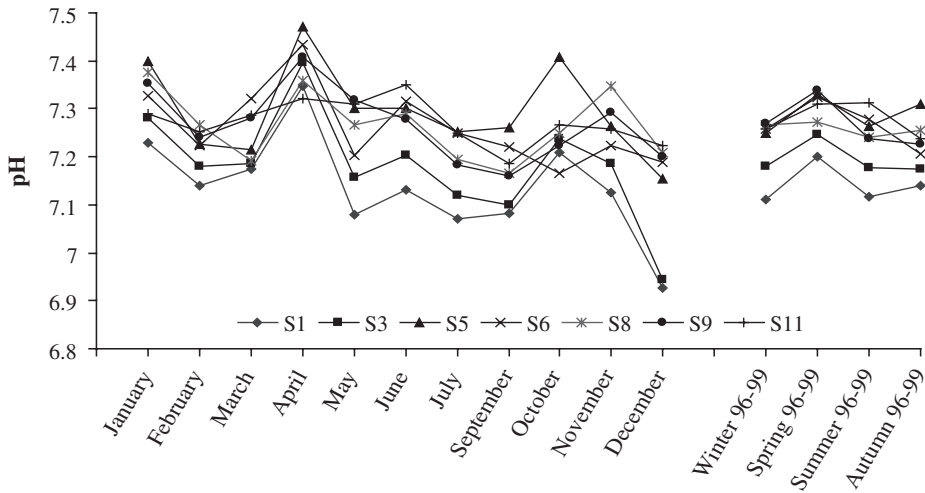


FIGURE 2 Monthly and seasonal trend of pH from selected station (S1, S3, S5, S6, S8, S9, S11) water samples collected between 1996 and 1999.

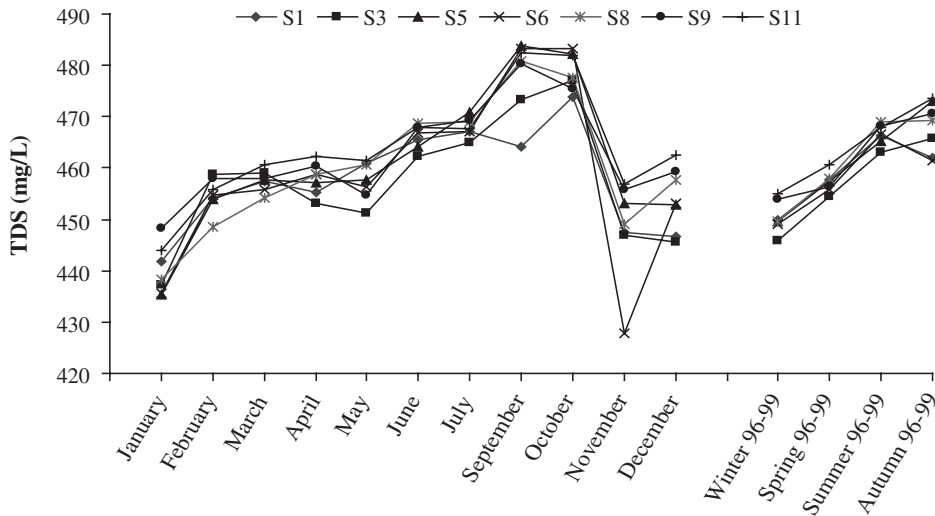


FIGURE 3 Monthly and seasonal trend of total dissolved solids (TDS, mg/L) from selected station (S1, S3, S5, S6, S8, S9, S11) water samples collected between 1996 and 1999.

TABLE III Spearman rank correlations (r_s) and significance levels between the microbiological properties of water samples collected at the S1, S3, S5, S6, S8, S9 and S11 sampling points during the period 1996–1999

	TC/FC	TC/FS	FC/FS
Winter	0.778 ^a	0.616 ^a	0.612 ^a
Spring	0.901 ^a	-0.004 ^b	-0.084 ^b
Summer	0.754 ^a	-0.122 ^b	-0.097 ^b
Autumn	1.000 ^a	0.507 ^a	0.507 ^a

^aStatistically significant for $p < 0.001$; ^bstatistically non-significant.

TABLE IV Spearman rank correlations (r_s) and significance levels between the microbiological and physicochemical properties of water samples collected at the S1, S3, S5, S6, S8, S9 and S11 sampling points during the period 1996–1999

	TC/T	TC/pH	TC/TDS	FC/T	FC/pH	FC/TDS	FS/T	FS/pH	FS/TDS
Winter	0.385 ^b	-0.393 ^a	0.120 ^d	0.359 ^b	-0.245 ^d	0.087 ^d	0.128 ^d	-0.297 ^c	-0.140 ^d
Spring	-0.137 ^d	-0.011 ^d	-0.058 ^d	-0.118 ^d	-0.057 ^d	0.108 ^d	-0.098 ^d	0.111 ^d	-0.038 ^d
Summer	0.067 ^d	-0.133 ^d	0.050 ^d	0.006 ^d	-0.010 ^d	0.042 ^d	-0.041 ^d	0.167 ^d	0.085 ^d
Autumn	-0.406 ^a	-0.351 ^b	-0.328 ^b	-0.406 ^a	-0.351 ^b	-0.328 ^b	-0.401 ^a	-0.297 ^c	-0.307 ^c

Statistically significant for ^a $p \leq 0.001$, ^b $p < 0.01$, ^c $p < 0.05$; ^dstatistically non-significant.

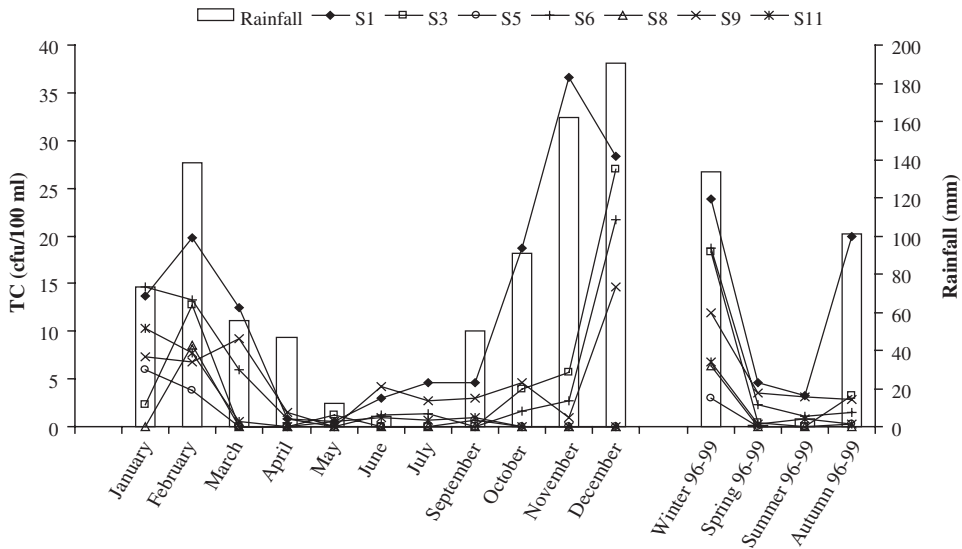


FIGURE 4 Monthly and seasonal trend of total coliforms (TC) from selected station (S1, S3, S5, S6, S8, S9, S11) water samples collected between 1996 and 1999.

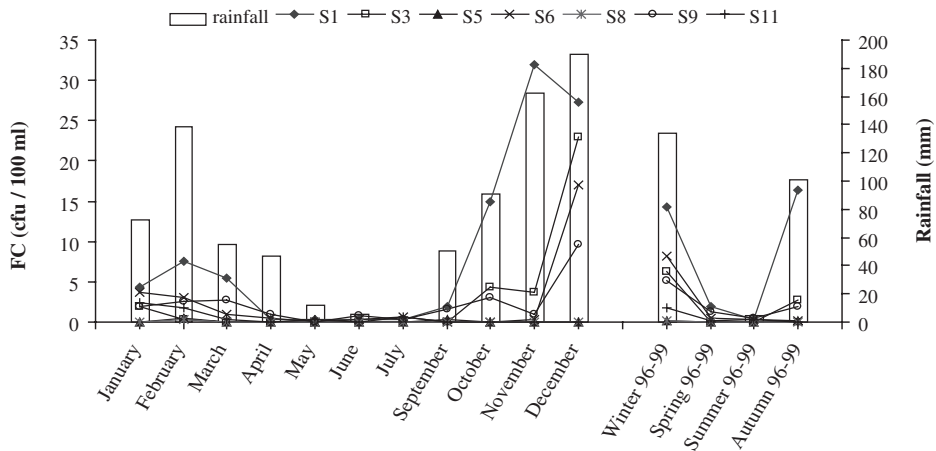


FIGURE 5 Monthly and seasonal trend of fecal coliforms (FC) from selected station (S1, S3, S5, S6, S8, S9, S11) water samples collected between 1996 and 1999.

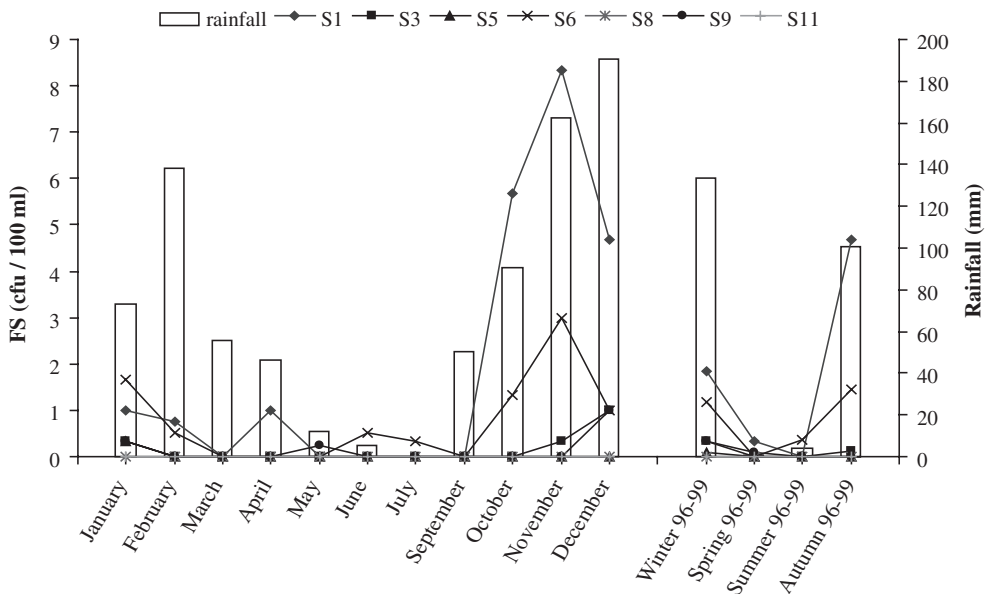


FIGURE 6 Monthly and seasonal trend of fecal Streptococci (FS) from selected station (S1, S3, S5, S6, S8, S9, S11) water samples collected between 1996 and 1999.

Failures for coliforms were much greater (more than double) during the autumn–winter period. A similar trend for coliforms has been reported elsewhere [13, 24].

In all seasons, TC was the indicator that exceeded the water quality standards most frequently (Fig. 7). During the winter study all the standard failures for any of the indicators reached 61%; 10% counts for TC alone, 46% included both FC and TC failure, 26% included both FC and FS failure and 21% failed for all three indicators. During the spring TC indicator failures were encountered for 28% of the samples (combined failures were 30% of the samples). In particular, 23% failed for both TC and FC indicators, 5% involved TC alone while 2% exceeded only the FS indicator.

The combined failures during summer were 30 and 26% included a TC failure. The summer pattern was similar to the spring one with 18, 8 and 4% of the samples, failing for both TC and FC, TC alone and FS alone respectively. Finally, during autumn 46% of the examined samples failed the quality standards; 43% included a TC failure, but 5% of the samples failed for TC alone. Moreover, 35% of the samples exceeded both TC and FC indicators, 16% exceeded both FC and FS indicators, 3% were for FS alone while 13% included failing for all three indicators.

It is worth noting the similar patterns observed during the autumn–winter period (wet weather) and the spring–summer (dry weather) period. During wet weather, there was much greater concordance among failures by the three indicators, as evidenced by the overlap in Fig. 7. This is further supported by the relatively higher Spearman correlations found among the three bacterial indicators during the autumn–winter period. During dry weather there was poor correlation among failures of the water quality standards. The periodic failure indicates two possible reasons for the poor quality of the supplies. Firstly, (and less probably) the source of contamination may itself be episodic and secondly that it may be due to various factors capable of introducing a seasonal dependence into the failure rate, which may include some combination of

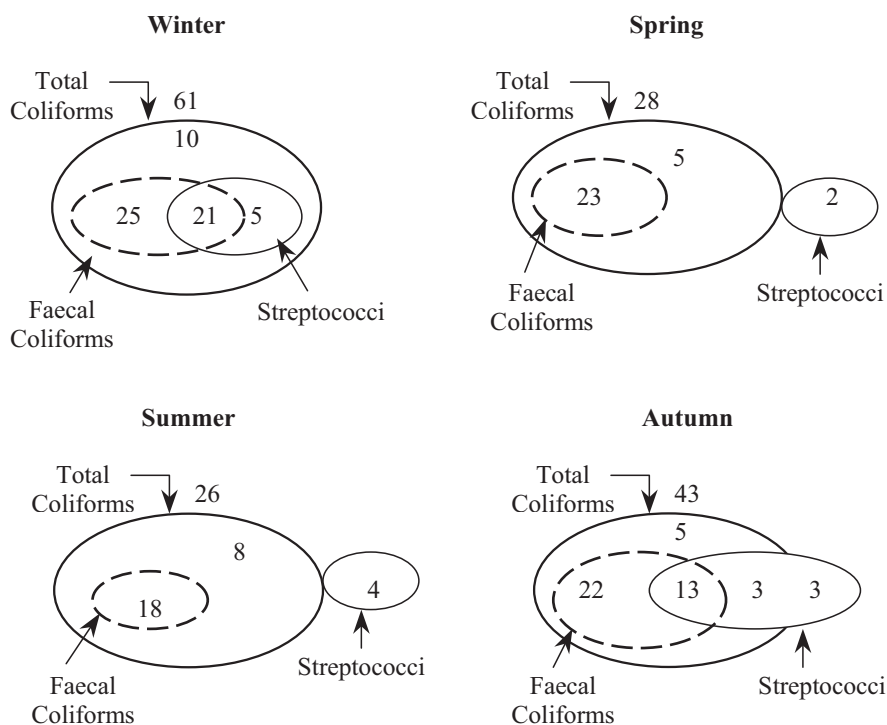


FIGURE 7 Percentage of microbiological indicator failures by indicator and season.

climatic, land management and hydrological factors. These considerations have implications for the success of management strategies aimed at improving source protection.

Comparing the failure rates between samples collected directly from the springs (S1) and at the end of the network pipeline (S5, S8, S11) in Table III can help to distinguish between these contaminant origins. The higher failure rate for samples collected directly from the springs compared to those taken from the potable tap suggests that the groundwater source itself contributes much of the microbiological load rather than the storage or supply-line contamination mechanism. Springs usually become contaminated when sewers, septic tanks, cesspools or other sources of pollution are located on higher adjacent land. However, in limestone formations, as in our study, contaminated material frequently enters the water-bearing channels through sinkholes or other large openings and may be carried along with groundwater for long distances. Nevertheless, when comparing the failure rates after disinfection with chlorine (S4, S7, S10) and at the end of the pipelines (S5, S8, S11) and especially at station S11 (city of Lefkada) there is evidence that the distribution network also affects the water quality to a certain extent. In drinking-water distribution systems, the occurrence of soft deposits and microbial biofilm can be associated with technical and hygiene problems. Microbes growing in biofilm reduce the water quality and can cause bad taste and odor. The accumulation of microorganisms on the pipeline surfaces and the formation of biofilm depend on many factors prevailing in the water system, e.g., type of surface material, microbial

occurrence in water, concentrations of nutrients and disinfectants, temperature and the hydraulics of the system [8, 25, 26]. Accidental contamination of the drinking-water network or inadequate disinfection are generally the reasons for the penetration of bacteria into a drinking-water distribution network. Free chlorine had no effect on the microbial numbers in the soft deposits and bacteria attaching to surfaces may increase their resistance against disinfection [27, 28]. Pipe materials can also affect the efficiency of a disinfectant. Holden *et al.* [26] have reported that free chlorine is an effective biocide on polyethylene whereas chloramine is effective on cast-iron surfaces.

CONCLUSIONS

Approximately 38% of the samples collected routinely along the water supplies of Arta, Preveza and Lefkada cities during 1996–1999 failed current drinking-water guidelines for coliforms. There was greater consistency among failures of bacterial indicator standards during wet weather (autumn and winter) than during dry weather (spring and summer). Although this observation is likely to be due to a combination of local and regional scale factors the variability was partially explained by changes in hydrological conditions (rainfall amount). Total coliform was the bacterial indicator that exceeded single sample standards most often regardless of the season of sampling. The higher failure rate for samples collected directly from the springs compared with those taken from the potable tap indicates that the groundwater source itself contributes much of the microbiological load rather than the storage or supply-line contamination mechanism. Finally, it is possible to suggest that a more efficient use of local available water resources can be achieved if periods of greatest contamination risk are identified and targeted for sampling. A microbiological risk-assessment survey in conjunction with an ongoing program of periodic monitoring of supplies for indicator organisms, could minimize the current risks by decreasing the potential for contamination of the water supplies at source.

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