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Contamination of many sources and irrigation canals by fecal bacteria in the piedmont of the middle atlas of beni mellal (Morocco), and physicochemical characteristics accompanying this contamination

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ABSTRACT

With a geologic structure which facilitates the emergence of numerous springs supplying in drinkable waters several urban and rural towns and irrigating the neighboring lands, the piedmont of the mountain range of the city of Beni Mellal is situated between the plain of Tadla in the northwest and the alignment of the synclines of Oued Abid in the Southeast. These sources are a little or not maintained at all; so they are exposed to various pollutions in particular those microbiological or physicochemical. In this work we have estimated the degree of contamination of eleven springs which are geographically distributed along a distance of 70 km. The physicochemical characteristics accompanying this infection were also determined. The results show that the majority of these sources or canals irrigation present fecal bacteria germs and the degree of the contamination is high in some of them. This contamination has an anthropological or animal origin in particular the use of fertilizer under the form of 'manure' and the excrements of cattle drinking or grazing near these sources. Besides, the physicochemical characteristics of the most polluted sources were different of the less polluted ones. Indeed, these last ones have relatively neutral pH, clear waters except those of Asserdoune, a low oxidability, an important hardness and alkalinity what is the opposite for the sources or the most polluted water of the seguias (irrigation canals).

KEYWORDS

Water sources;
Fecal bacteria;
Physicochemical;
Middle Atlas;
Morocco.

Regular Paper

INTRODUCTION

The water is an element of life and a tool for economic development of the humanity. However, the Man, by diverse industrial, agricultural and mining activities degrade the quality of this substance causing so its pollution by physical, chemical or biological. As example of the biological pollution is the contamination of the environment with germs of pathogenic bacteria, in particular those of fecal origin, by rejecting untreated waters from sewer into the natural aquatic environment, causing problems to human health like typhoid, cholera, etc.

Besides, the Who (O.M.S., 1992)[1] considers that the poor microbiological quality of drinkable waters remains the first cause of health problems in the world. Germs responsible for these diseases are transmitted to water by faecal matter. For these reasons, the bacteria which are exclusively found in high densities in the feces are used as indicators of a faecal pollution and their presence in the water indicates a risk of a biological contamination. So, the faecal coliforms and the fecal enterococcus are considered as indicator bacteria of the water faecal contamination. As a consequence, the detection of faecal coliforms in the water warns us about a possible contamination of faecal origin[2] Also, according to Zmirou and *al*. (1987)[3], and Edberg and *al*. (2000)[4], the faecal coliforms Escherichia coli and the enterococcus, are more valid such as risk indicators. In gross water at high concentrations of thermotolerant coliforms (including E. coli) the probability to notice the presence of enteric protozoa and of enteric viruses was also very high[5,6].

The monitoring of the water microbio-logical quality thus is a means of control of the conservation of the public health. The microbiological control of the water is mainly based on the research of indicator bacteria of fecal contamination. But, it is necessary to note that these bacteria have not always a pathogenic character; but, their presence indicates the existence of a fecal contamination and thus an epidemiological risk.

In this work we have studied the microbiological contamination, during a period situated between 09/2006 and 06/2007, of eleven sources situated on the piedmont of the Atlas Mountains of Beni Mellal (Morocco) (TABLE 1). In every one of them, we have seasonally evaluated the concentrations of three hygienic indicators (total coliforms, faecal coliforms, and fecal streptococci).

MATERIAL AND METHODOLOGY

The studied zone (Figure 1), called piedmont of Atlas of Beni Mellal, is situated between the plain of Tadla on the northwest and the alignment of the synclines of Oued Abid on the South. It covers a bande along Tadla chain called Atlas of Beni Mellal[3,4]. The mountain range of piedmont shows summits reaching 2411 m at Jbel Ighnyme and 2248 m at Jbel Tacemit. The geology of the area is dominated by rocks of the liasic period and that are essentially of carbonated nature. This geological formation has developed a superficial and subterranean karstic bed allowing the emergence of numerous karstic sources along the fault of Tadla. The most important one is Ain Asserdoune which is considered as one of the most important karstic springs in Morocco[7] and which constitutes the main source of drinkable water supply of an urban area of more than 163 248 inhabitants: the city of Beni Mellal.

In this work we have estimated, in the piedmont of Beni Mellal, the bacteriological contamination degree according to seasons in eleven sources and in four sites

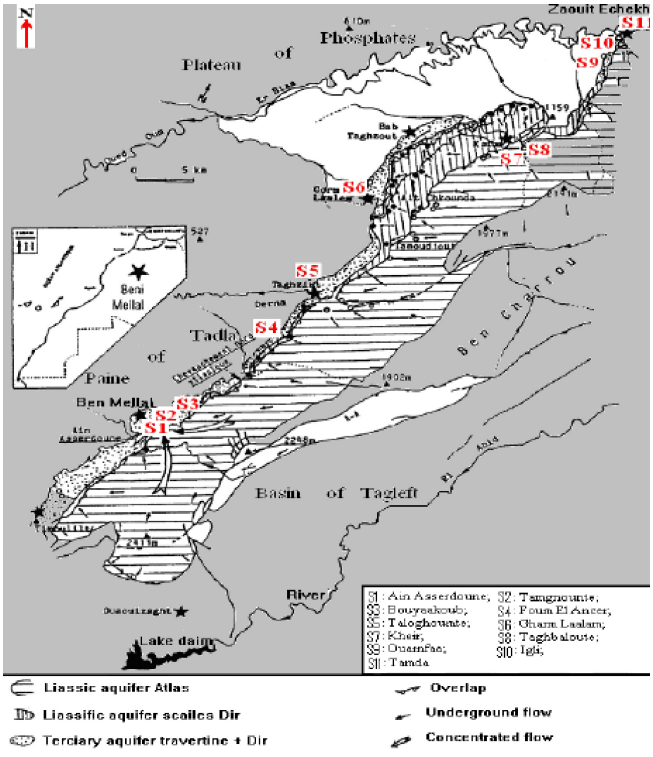

Figure 1 : Hydrogeological map of the Atlas of Beni Mellal (Bouchaou, 1995)

Regular Paper

TABLE 1 : Given codes to the samples according their source and their season

Source	Samling season	Code	Source	Samling season	Code
	Summer	OE		Summer	BE
Ouarnfaa	Fall	OA	Bouyakoub	Fall	BA
	Winter	OH		Winter	BH
	Spring	OP		Spring	BP
	Summer	IE		Summer	AE
Igli	Fall	IA	Asserdoune	Fall	AA
	Spring	IP		Winter	AH
	Summer	TE		Spring	AP
Tamda	Fall	TA		Summer	BVE
	Spring	TP	Belle Vue	Full	BVA
	Summer	TgE		Winter	BVH
Taghbaloute	Fall	TgA		Spring	BVP
	Spring	TgP		Summer	OAE
	Summer	KE	Ouled Ayad	Fall	OAA
Ksiba	Fall	KA		Winter	OAH
	Spring	KP		Spring	OAP
	Summer	GE		Summer	TmE
Gharm Laalam	Fall	GA	Tamgnounte	Fall	TmA
	Winter	GH		Winter	TmH
	Spring	GP		Spring	TmP
	Summer	TlE		Summer	ME
Taloghomte	Fall	TlA	Mdiwla	Fall	MA
	Winter	TlH		Winter	MH
	Spring	TlP		Spring	MP
	Summer	FE		Summer	OHE
Foum el Ancer	Fall	FA	Ouled Hamdan	Fall	OHA
	Winter	FH		Winter	OHH
	Spring	FP		Spring	OHP

TABLE 2 : Eigenvalues and contribution percentages of the three first axes

Axis	Eigenvalues	Inertia rate (%)	Cumulative variance (%)
C1	5.70	31.7	31.7
C2	3.26	18.1	49.8
C3	1.98	11	60.8

situated on some canals of irrigation that are locally called "seguia" ; Two sites are located along a seguia of Ain Asserdoune (Belle vue station and Ouled ayad station) and two other ones are located along Ain Tamgnounte (Mdiwla station and Ouled hamdan station).

So, we have evaluated the content of the total coliforms (CT), faecal coliforms (CF) and faecal streptococci (SF) in these sites or sources. The water of those sources is used to drink and to the cultures irrigation. The sources are spread over a distance of 70 km separating the city of Beni Mellal and the city of Zaouit Cheikh (Figure 1) and every sample Analyzed was codified according to TABLE 1.

Bacteriological analysis

The counting of the total coliforms (CT), of the fecal coliformes (CF) and of the fecal streptococci (SF), was realized according to the method of the most probable number (MPN)[8,9]. This technique consists in spreading a volume of sample in a certain number of tubes and to look for the presence or the absence of the studied germs, the results are given by the tables of Mac Grady allowing the calculation of MPN[10]. The collected samples are put in 500 ml glass flasks previously sterilized in the autoclave, then transported in an icebox at the temperature of 4°C in the analysis labora-

tory. The used milieu and the incubation conditions : for the coliforms; lauryl milieu, after an incubation of 48 h at 37°C (presumptive test), the "positive" tubes (fermentation of the lactose and production of gas) are transplanted for a confirmative test in a selective milieu containing biliary salts BLBVB (Broth of Biliary Lactose with Brilliant Green) incubated during 48 h at 37°C (total coliforms (CT)) and EC milieu broth for the fecal coliforms CF incubated during 48 h at 44°C. Concerning the streptococci, their research is made on Rothe milieu at 37°C during 48 h (presumptive test). From the positive Rothe tubes, we make a transplanting on Litsky milieu at 37°C during 48 h.

RESULTS AND DISCUSSION

Presence of total coliforms and of fecal coliforms in the studied sources

The TABLE 3 illustrates the bacteriological loads estimated in the various studied sources. According to the results, the majority of these sources show the presence of total coliforms and of fecal coliforms. The most important bacterial load (1100 CT/100 ml and 240 CF/100 ml) were recorded in summer in Tamgnounte source; also, the autumnal values of Ouarnfaa source (460 CT/100ml and 290/100 ml) were imprtant. Let us note that the water of Tamgnounte source is used for drinking to cattle, for washing the linen; also, it is used for swimming when it is hot. All this activities constitute a potential origin of this bacteria contamination. The strong values of the Ouarnfaa's source can be explained by its situation downstream domestic waste waters of the urban area of the city of Zaouit Cheikh.

TABLE 3 : Distribution of the contribution of the 15 variables and PCA code (Correlation between the variables and the principal axis)

Variables	Code	Percentage of inertia explicated		
		Axis C1	Axis C2	Axis C3
Total Coliforms	CT	0.8036	-0.1212	-0.2053
Fecal Coliforms	CF	0.7679	-0.0997	-0.1700
Fecal Streptococci	SF	0.6653	-0.0577	-0.0642
Temperature	Tp	0.1828	0.4471	-0. 4917
pH	Ph	0,8843	0.0749	0.0449
Conductivity	Cd	- 0.3466	0.7796	-0.1399
Turbidity	Tr	0.6305	0.1323	0.0316
Dissolved Oxygen	OX	0.4279	-0.3811	-0.5339
Oxidability	Ot	0.7005	0.4727	0.1677
Hydrometric rate	TH	-0.6709	0.5682	0.0368
Complete Alcalimetric rate	TAC	-0.5941	0.3600	-0.3568
Calcium	Ca	-0.3484	0.4691	0.6393
Magnésium	Mg	-0.4019	0.1121	-0.6716
Nitrate	N3	-0.3465	0.2562	-0.4820
Nitrite	N2	0.2805	0.5612	0.1378
Ammonium	NH4	0.5229	0.4019	0.1837
Chloride	Cl	0.4807	0.5840	0.0148
Sulfate	SO	0.4787	0.6835	-0.2345

At the level of the springs of Igli, Gharm Laalam and Bouyakoub there is always a presence of germs but with concentrations lower than the evoked sources. The recorded values vary between (15-93/100 ml) for the total coliforms and (9-93/100 ml) for the fecal coliforms.

The concentration found at the levels of the sources of Tamda, Taghbaloute, Ksiba and Foum El Ancer are very low even seasonally null. This low contamination could be explained by their long distance from inhabited areas. Indeed, all these sources are located upstream of the cities.

In Ain Asserdoune source, which constitutes the main source of drinking water of the Beni Mellal city, we noted a strong autumnal value of germs (on 1110 CT/100 ml and 210 CF/100 ml) just after the fall of rain. Probably the infiltration of the superficial water in the aquifer could be an origin of this important contamination.

For the fecal streptococci, the high values were recorded at the level of Gharm Laalam and Asserdoune (93 SF/100 ml) and at the Taghbaloute springs (43 SF/100 ml). At the contrary, the contamination is less important in the other sources. The contamination of these springs is explained by the existence of lost wells receiving fecal matter and also excrements of cattle grazing in the neighborhood farmlands close to the sources.

Influence of some physical and chemical parameters of the environment and bacterial contamination, typological analysis:

With the aim of characterizing in a physico-chemical way the conditions that accompany the infestation

Regular Paper

of the studied sources by pathological germs of fecal origin (CT, CF and SF), we established a physico-chemical typology of the surveyed environments by applying the Principal Component Analysis (P.C.A) to all the physico-chemical data (15 parameters) and bacteriological (3 groups of indicator germs of fecal contamination: CT, CF and SF) collected. Let us remind that the PCA is a factorial statistical method allowing to simplify the description of a table containing lots of measures of many variables which have quantitative aspect, coming from a time-space observation plan and that are difficult to approach[11]. This statistic method is widely used in biology field such as the work of Berrady and *al*. (2000)[12].

The final matrix of data analyzed is constituted of a double entry table of 18 variables (columns) x 56 samples (lines). The results (Tab. 2) show that the first axis (P1) extracts 31.7% of the explained inertia.

The second (P2) 18.1% and the third axis (P3) 11%, i.e. a total of 60.8%, a total that is enough for the interpretation of the plan P1xP2 and P1xP3. The contributions of the various variables in the constitution of the first three axes are illustrated in the TABLE 3.

Interpretation of the axis P1, P2 and P3

Component P1

The analysis of the results (TABLE 3, and Figure 2) shows that the variables which participate significantly in the constitution of the axis P1 are the pH (0.8843%), the Turbidity (0.6305%), the Oxidability (0.7005%), the ammoniacal nitrogen (0.5229%), the Hydrometric rate (0.6709%) and the complete alkalimetric rate TAP (0.5941%). From the negative side to the positive side of P1, we have noted an increasing gradient of the pH, Tr, Od, NH4+ and a decreasing gradient of the

TAC and TH. According to the nature of the variables which contribute significantly on this axis, thus it is an axis representing an increasing organic pollution from the negative side to the positive side of the axis P1.

Moreover, the Figure 2 shows that the high concentrations of the CT (0.8036%), CF (0.7679%) and SF (0.6653%), are situated in the positive part of the axis P1. So; the ecological conditions which seem to facilitate the reproduction of these germs are characterized by high concentrations in NH4+, Od, Tr and by an alkaline pH.

Component P2

For the axis C2, TABLE 3 and Figure 2 show that only four variables contribute significantly in its constitution, i.e. Cd (0.7796%), N2 (0.5612%), Cl (0.5840%), SO (0.6835%) and which are all situated in the positive part of this axis. The nature of these variables correlating to C2 shows that this last one corresponds to a gradient of mineralization which increases from the negative pole to the positive pole of the same axis.

Component P3

The variables which present a significant correlation with the axis C3 (TABLE 3 and Figure 3) are the Temperature (0.4917%), dissolved oxygen (0.5339%), magnesium (0.6716%), nitrates (0.4820%), and calcium (0.6393%). The first four variables are all situated in the negative part of the plan; only the calcium is situated in the positive side. So, this axis represents an in-

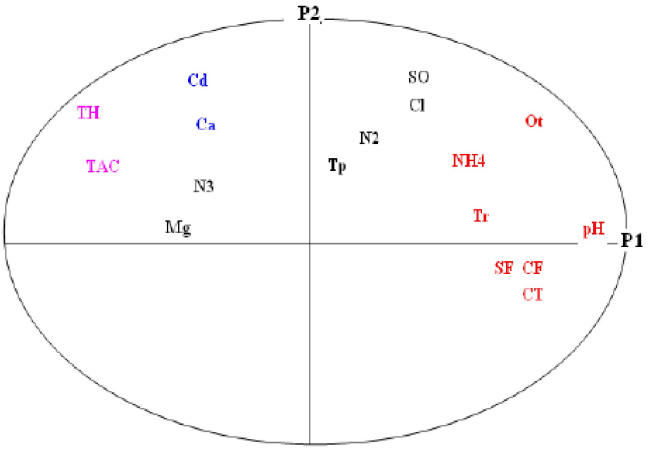

Figure 2 : Projection of the physico-chemical and bacteriological variables in the plan 'P1xP2

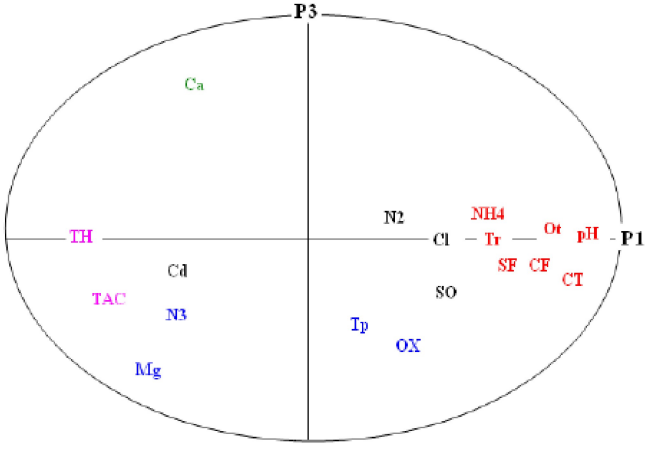

Figure 3 : Projection of the physico-chemical and bacteriological variables in 'P1 xP3' plan

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creasing gradient for the calcium concentration and a decreasing gradient for the temperature values, and the oxygen, magnesium, and the nitrates concentrations.

Analysis of the plan P1xP2

The projection of points representing the samples on the plan P1xP2 (Figure 4) shows a differentiation of two groups:

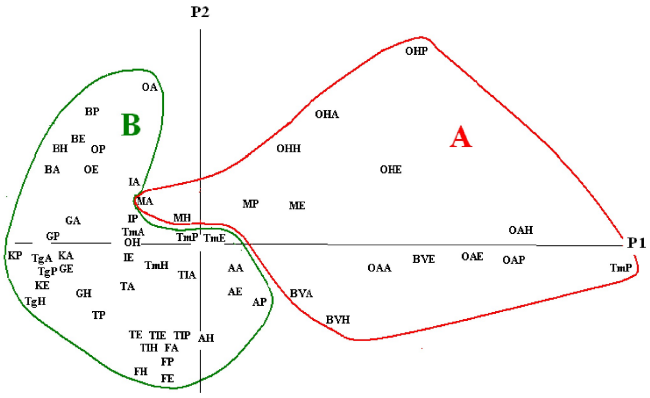

Figure 4 : Projection of the points representing the samples on the plan P1x P2

A first group of samples, called A. All these samples were made in some stations presenting a wealth in organic matters, an alkaline pH, the high values in turbidity and nitrite, and the low contents in total hardness and in TAC. These physico-chemical conditions are better realized in the stations called Belle Vue, Ouled Ayad, Mdiwla and Ouled hamdan which all situated in the irrigation canals (seguias) issued from Asserdoune and Tamgnounte sources with the exception of a station that was located in Tamgnout source itself.

Besides, the stations where the samples of the group A were made are characterized by a high annual abundance in total coliforms, fecal coliforms and fecal streptococci. The origin of this strong bacterial contamination is maybe linked to the strong domestic activity (wash of the dishes or the linen, the direct refusal of the garbage, drinking point for animals) which enriches the environment in organic matter and in nutrients. These last ones constitute nutritive sources essential for the survival and the reproduction of bacteria.

A second group of samples, called B, corresponding to some sources presenting a less important degree of contamination in coliforms and enterococcus and which are physico-chemically characterized by relatively neutral pH, clear waters except those of Asserdoune, a low oxidability, an important hardness and alkalinity Moreover, the Figure 4 shows that some levies are situated on the positive side of P2. So, according to the physico-chemical meaning of this axis, these levies are more mineralized than those illustrated in the other side of P2. Indeed, the sources of these levies have a high values in chloride, sulfate and the conductivity. Of the first group of sources or sites we quote those called Bouyakoub, Ouarnfaa, Mdiwla and Ouled Hamdan. But in P1xP2 plan (Figure 4), the points representing the variables FC, ST and SF all occupy a central position of the axis P2. This suggests that there are no particular physicochemical conditions that accompany the abundance of these organisms according to the physicochemical parameters that contribute significantly in the constitution of P2.

Analysis of the plan P1xP3

Furthermore, the projection plane P1xP3 (Fig-

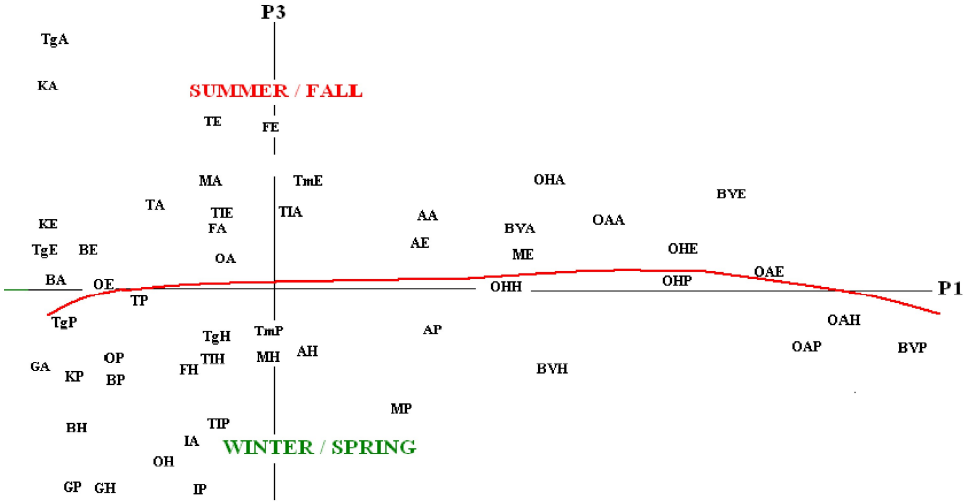

Figure 5 : Projection of the points representing the samples on the plan P1x P3

Regular Paper

ure 5), highlights the effect of the 'season' as a factor influencing the water physico-chemical characteristics.

Indeed, the samples taken in summer and autumn are grouped at the positive side of the axis P3, and are characterized by the high concentrations of calcium, low concentrations of magnesium and nitrate, and low temperatures values. In contrast, the Figure 4 shows that the points representing the levies conducted in winter or spring are located in the negative side of the axis P3, and are characterized by high concentrations of magnesium, nitrate and oxygen, a low concentration of calcium, and a relatively high temperature.

Concerning the abundance of the fecal bacteria, the TABLE 4 shows that this abundance was not depending to the physicochemical parameters that contribute to the formation of the P3 axis. The importance of the abundance values recorded in summer or fall on one hand and winter or spring on the other hand does not depend on these periods. The abundances of these organisms in sites on seguia (Belle Vue, Ouled Ayad, Mdiwla and Ouled Hamdan), ie the most polluted sites are good examples

TABLE 4: Infestation rates of different sites by the CF, CT and SF

Germes→ Site↓		CT	CF	SF	Germes→ Site↓		CT	CF	SF
	Summer	290	290	9		Summer	15	15	7
Ouarnfaa	Autumn	460	240	93	Bouyakoub	Autumn	23	0	0
	Winter	43	43	4		Winter	0	0	0
	Sprins	43	43	4		Sprins	15	9	0
	Summer	23	23	7		Summer	0	0	0
Igli	Autumn	43	4	0	Asserdoune	Autumn	1100	210	93
	Winter	0	0	0		Winter	0	0	0
	Sprins	23	23	7		Sprins	4	3	4
	Summer	0	0	0		Summer	160000	75000	39000
Tamda	Autumn	14	9	0	Belle Vue	Autumn	46000	15000	4300
	Winter	0	0	0		Winter	110000	46000	900
	Sprins	0	0	0		Sprins	1100000	460000	240000
	Summer	0	0	0		Summer	210000	150000	93000
Taghbalout	Autumn	4	0	43	Ouled Ayad	Autumn	210000	75000	46000
	Winter	0	0	0		Winter	460000	29000	4300
	Sprins	0	0	0		Sprins	460000	210000	2800
	Summer	4	4	0		Summer	1100	1100	7
Ksiba	Autumn	9	0	0	Tamgnount	Autumn	2	0	4
	Winter	0	0	0		Winter	4	0	15
	Sprins	0	0	0		Sprins	93	15	0
	Summer	23	23	0		Summer	160000	120000	75000
Gharm	Autumn	93	93	4	Mdiwla	Autumn	46000	4600	1100
Laalam	Winter	9	0	3		Winter	15000	2100	2800
	Sprins	43	43	93		Sprins	110000	15000	430
	Summer	43	43	21		Summer	290000	150000	93000
Taloghomt	Autumn	150	75	7	Ouled	Autumn	46000	24000	46000
	Winter	0	0	4	Hamdan	Winter	46000	15000	1500
	Sprins	4	4	4		Sprins	120000	23000	1200
	Summer	0	0	0	-	-	-	-	-
Foum	Autumn	0	0	0	-	-	-	-	-
Ancer	Winter	3	0	15	-	-	-	-	-
	Sprins	0	0	0	-	-	-	-	-

CONCLUSION

The contamination by the germs of fecal origin and of waters of certain springs and seguias is very high. The anthropological activity represented by a domestic activity, a rejection of liquid or solid waste in the unprotected sources seems to be the main cause of this bacteriological pollution. The spreading of fertilizers under the form of manure, the rejection of excrements of the cattle grazing on the farmlands or watering the water of these water sources are also potential causes of this contamination. So, the protection of these resources is necessary for the support of the biological quality of waters of the prospected springs. Concerning the physico-chemical conditions which characterized these contaminated springs and seguia we noted: a wealth in organic matters, alkaline pH, high values of turbidity and nitrite and low contents in total hardness and in TAC.

Besides, the springs that are less or not contaminated by fecal bacteriological germs have shown the same physico-chemical conditions of those that are more contaminated. They are characterized by relatively neutral pH, clear waters except those of Asserdoune, a low oxidability, hardness and a quite important alkalinity.

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