

## Quality and disinfection trials of consumption water in storage reservoirs for rural area in the Marrakech region (Assif El Mal)

Faissal Aziz, Laila Mandi, Abdellatif Boussaid, Fatima Boraam and Naaila Ouazzani

### ABSTRACT

Traditional reservoirs for water storage are important systems of water supply in rural areas of Morocco. These reservoirs are fed by rainwater and/or directly from rivers through open channels; the stored water is used without any treatment as drinking water by the surrounding population. The present study aimed to assess the physicochemical and bacteriological quality of stored water and the corresponding sediment in six traditional reservoirs (R1 to R6) located in the rural municipality of Assif El Mal. We tested two inexpensive methods of disinfecting the stored water: chlorination and solar disinfection in bottles. The results show a rise of organic and mineral concentrations. Regarding bacteriological quality, a critical contamination level was detected ( $8 \times 10^5$  CFU/100 ml in water and  $9 \times 10^7$  CFU/g in sediment) according to the 2002 Moroccan Standards for drinking water (0 CFU/100 ml). In the disinfection tests, chlorine disinfection removed all studied germs after just 1 hour, and the solar exposure process removed the majority of bacteria (after 3 hours) except those with a resistant form (*Clostridia*).

**Key words** | bacteriological contamination, chlorination, drinking water, rural area, solar disinfection, storage reservoirs

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### INTRODUCTION

Morocco is a Mediterranean country facing water scarcity because of arid and semi-arid conditions. Consequently, various problems of development and sustainable management of water resources occur. This situation is exacerbated in rural areas as a result of the huge lack of appropriate infrastructure, namely drinking water and sanitary systems. This is the case in our study site, the Assif El Mal valley, which has a limited water potential, which makes it more difficult to obtain good water quality.

Often, the rural population uses a variety of sources of drinking water, including groundwater, rainwater and surface water. The last two water sources are collected in reservoirs for use when no other permanent drinking water sources are available. Assessment of the quality of these alternative water sources is necessary because of

their direct effect on people's health. Indeed, many studies have reported sanitary problems related to water quality in rainwater storage tanks (Taylor *et al.* 2000; Abbott *et al.* 2007). The poor quality of these waters and the lack of proper treatment can cause health and environmental problems. Caslake *et al.* (2004) noted that contaminated water causes 6–60 billion cases of gastrointestinal diseases annually, occurring mostly in rural areas of developing countries, and Hunter *et al.* (2002) estimated that water-borne diseases kill more than 5 million people each year. The pathogens responsible for most of these deaths originate from human and animal faeces.

Two proposed water treatments are chlorination (Rincon Benavides 2005) and solar disinfection (SODIS); the latter is generally recommended in countries with a

warm and sunny climate (Acra *et al.* 1990; Rojko 2003; Rincon Benavides 2005; McGuigan *et al.* 2006). Chlorine and its compounds are relatively inexpensive and are convenient because of their strong antibacterial effects, notably the destruction of enzymes necessary to the life of pathogens. In contact with water, chlorine and its compounds release the active hypochlorite ions that destroy most bacteria, viruses and parasites (Pascual 1998; Bouziani & Aslah 2009). SODIS eliminates a wide range of organic compounds, chemicals and pathogenic organisms by direct exposure to sunlight. This technique is relatively inexpensive, and avoids the generation of harmful by-products (Calkins *et al.* 1976), as well as being able to disinfect about a litre of water in 30 min (Caslake *et al.* 2004). It is also a point-of-use method which avoids secondary water contamination that commonly occurs through storage (Mintz *et al.* 2001).

Scientists have devoted a great deal of time and effort to investigate the quality of stored rainwater (Yaziz *et al.* 1989; Simmons *et al.* 2001; Evans *et al.* 2006, 2009). In contrast, few workers have made a comprehensive assessment of the quality of river water stored in traditional reservoirs and used for human consumption. Furthermore, even the disinfection of water by a simple process that might be applied by isolated populations has rarely been addressed. Studies on disinfection processes could not assess their effectiveness against those with a resistant form, i.e. spore-formers.

The main aims of the present study are: (1) to assess the physicochemical and bacteriological quality of drinking water stored in traditional reservoirs in the rural municipality of Assif El Mal; and (2) to study the possibility of water disinfection by two simple and inexpensive methods that are suitable for the local socio-economic conditions: chlorination and SODIS in bottles.

## MATERIALS AND METHODS

### Overview of study area

#### Geographical framework

The basin of Assif El Mal is located on the north side of the High Atlas, 100 km southwest of Marrakech. The area has

moderate relief, with an average altitude of 600 m, and becomes higher towards the south near the Atlas Chain. The Assif El Mal valley is in the watershed of Mejjate, which extends between the Nfis River and the border of the plateau of Essaouira. It covers an area of approximately 2,200 km<sup>2</sup>. It is a left-bank tributary of the Tensift River, which is the main river of the plain of Haouz (Figure 1(a)).

#### Hydrology

The Haouz plain benefits from its proximity to the High Atlas Mountains. This factor makes it a hydraulic reservoir because of the relative abundance of rainfall that it receives. Some precipitation is in the form of snow that falls from December to April. Starting from March, snow usually melts and recharges the rivers (Jolly 2000).

The regime of the Assif El Mal River is irregular over the course of the year. The flow remains low with a maximum in January, but during the summer months the river is almost dry. This forces the local population to store river water in traditional reservoirs during the winter, and then use it for the rest of the year.

#### Climate

The climate of the watershed of Assif El Mal is characterized by extreme aridity. Its intensity depends primarily on altitude; continentality has only a very small effect. Otherwise, the seasonal contrast is well marked with a hot summer and cold winter (annual minimum temperature = 17 °C; annual maximum temperature = 38 °C). Irregular, intense and violent rainfall is often concentrated during autumn and winter. For the rest of the year, a considerable amount of drought occurs, especially in lowland areas where the temperatures and evaporation are high.

#### Study area

In the Assif El Mal valley, the population living in the plains suffers from drinking water scarcity. In terms of hygiene conditions, the local populations do not respect their surroundings as household rubbish and faecal waste are disposed off anywhere.

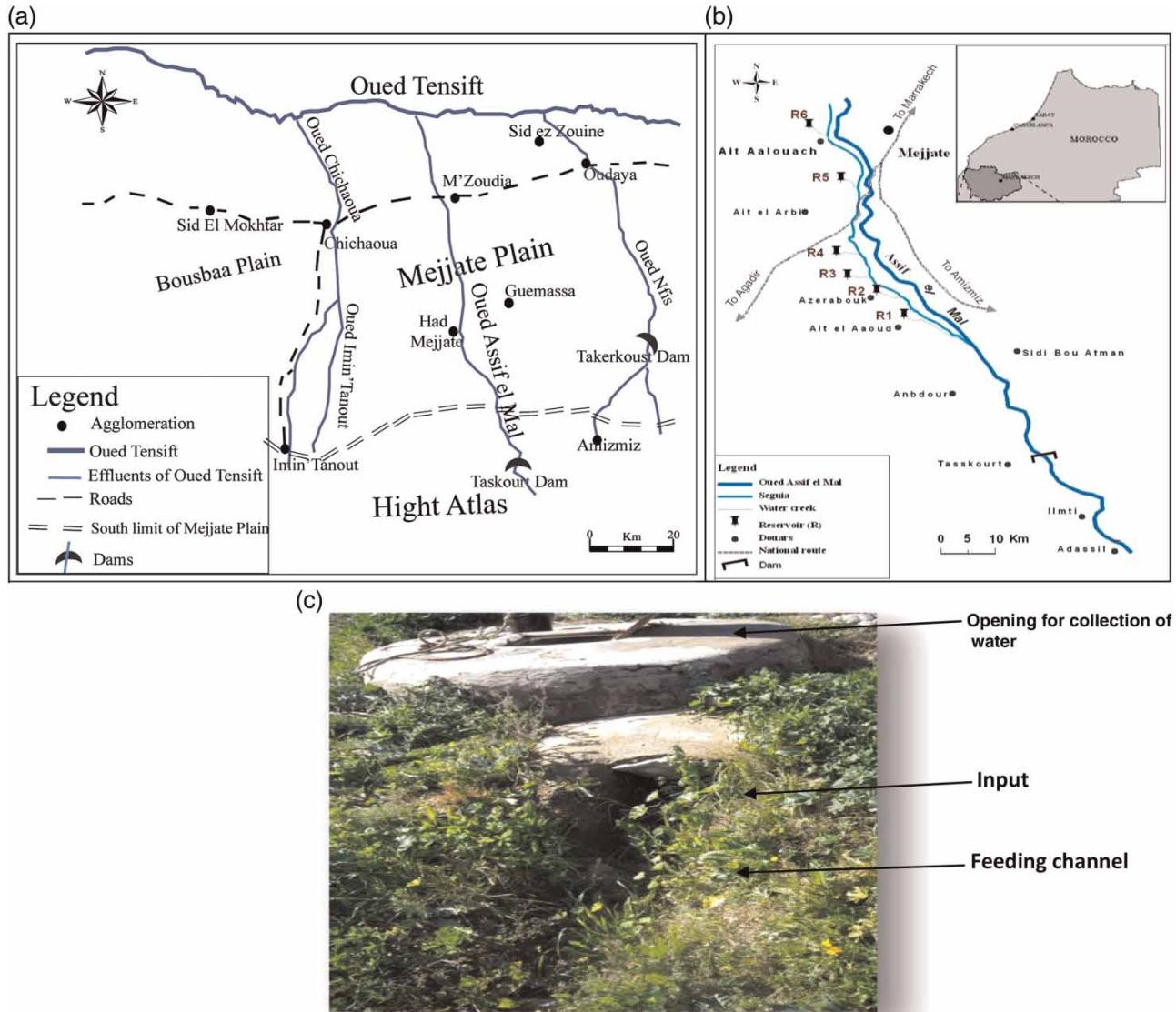


Figure 1 | (a) Map of the valley Assif El Mal situation; (b) location of sampling sites (R: traditional reservoir); (c) traditional reservoir.

The poor socio-economic status of the local people prevents them from digging wells. Consequently, water stored in the traditional way is their only source of water. This water is used for all purposes, including personal consumption and watering of livestock, without any prior treatment.

The traditional storage method uses reservoirs known locally as *matfya*. The *matfya* is a type of cistern, buried in the ground to a level of three-quarters of its total height. A *matfya* may be tubular in shape, 4–9 m in depth and 2–4 m in diameter, or rectangular in shape, 6–10 m in length, 3–5 m in width and

2–4 m in depth. These reservoirs are supplied by river and/or rainwater through channels called *segua* (Figure 1(c)). Table 1 describes the average characteristics of the water supplying these reservoirs. This water is likely to be contaminated by all activities conducted along the route of *seguias*.

In addition, the socio-cultural behaviour of the population towards water leads them to build more reservoirs than wells, as each family has a hereditary right to its share of surface water, whenever it is distributed along the *seguias* and stored in reservoirs for its own consumption.

**Table 1** | Average physicochemical and bacteriological characteristics and standard deviations of the Assif El Mal river waters supplying the reservoirs studied, during 2010 and 2011 ( $n = 16$ )

Physicochemical characteristics		Bacteriological characteristics	
Parameter	Value	Bacteria	CFU/100 ml
pH	7.7 ± 0.05	Faecal coliforms	250 ± 12
Conductivity (mg/l)	494 ± 22	Total coliforms	1,084 ± 56
Chloride (mg/l)	11.04 ± 3	<i>Escherichia coli</i>	73 ± 9
Bicarbonate (mg/l)	2.7 ± 0.8	Streptococci D	174 ± 22
Calcium (mg/l)	10.5 ± 2	Intestinal enterococci	234 ± 46
Sulphate (mg/l)	18 ± 1.7	<i>Staphylococcus aureus</i>	347 ± 32
Magnesium (mg/l)	142 ± 16	Clostridia (SRA)	855 ± 44
Oxygen (mg/l)	6.86 ± 0.64	<i>Salmonella</i> spp.	–
BOD <sub>5</sub> (mg/l)	4.15 ± 0.5	<i>Pseudomonas aeruginosa</i>	–
Nitrite (mg/l)	–		
Oxidability by KMnO <sub>4</sub> (mg/l)	2.04 ± 0.5		

## Sampling

Water and sediments samples were collected from six traditional reservoirs, which stored water intended for human use (Figure 1(b)). Sampling was according the French standard methods (AFNOR 1997) and Rodier (1996). The study reservoirs were chosen according to four main characteristics: urbanization, intensity of usage, accessibility and spatial distribution along the valley.

The water samples were taken monthly during winter (January–March) and summer (June–August) 2010, and were collected using plastic bottles for chemical assays, and in sterile glass bottles for bacteriological studies. Sediment samples were taken once in winter and once in summer from the same reservoirs as the water samples using a Shipek grab. Sediment samples were stored in sterile plastic bags.

All the samples were transported to the laboratory in the dark in a cooler at a temperature of 4 °C.

## Parameters analysed and methods used

### Physicochemical parameters

For water samples, temperature, pH, electrical conductivity and dissolved oxygen were measured *in situ* using a multi-parameter probe type WTW LF 92. The mineral nutrients (nitrite, sulphate, chloride, bicarbonate, calcium, magnesium, phosphorus) and 5-day biochemical oxygen

demand (BOD<sub>5</sub>) data were measured in the laboratory by spectrometry or volumetry according to French standard methods (AFNOR 1997) and Rodier (1996).

Sediment samples were first dried at 105 °C for 24 hours and then sieved to 2 mm. The main parameters analysed for sediment were as follows:

- *Total limestone*: the total content of limestone was determined by volumetric acidimetry with a Bernard calcimeter (AFNOR X31-103).
- *Organic matter*: organic carbon was determined by the modified Walkley & Black (1934) method.
- *Total nitrogen*: nitrogen was determined by Kjeldahl mineralization and distillation of ammonium and a final acidimetric titration.
- *Total phosphorus*: phosphorus was determined using an automated molybdenum-blue colorimetric procedure, following a sulphuric-peroxide digestion and pH adjustment.

### Microbiological analysis

The methods were quantitative for the indicator organisms of faecal contamination and qualitative for pathogens (Moroccan Standards 2006).

Membrane filtration was used to enumerate faecal coliforms (FC) and total coliforms (TC), Streptococci D (SD), *Escherichia coli* (EC), intestinal enterococci (IE), *Staphylococcus aureus* and *Pseudomonas aeruginosa*. The most

probable number (MPN) technique was used for water samples suspected to be highly contaminated by the same pathogens.

The culture media used were as follows:

- Lactose TTC agar with Tergitol, for coliform counts, and soybean casein digest agar for *E. coli* (Moroccan Standard 08.0.124).
- Bile aesculin agar for Group D streptococci counts and Slanetz and Bartley medium for enumeration of IE (Moroccan Standard 03.7.001).
- Baird Parker agar (BP-Agar) was used to select *S. aureus*. Identification was confirmed by growing the strains on beef-heart infusion agar followed by a coagulase test with rabbit plasma) (Moroccan Standard ISO 6888-1-2008).
- Sulphite-reducing anaerobes (SRA) were counted on SPS agar (sulphite polymyxin sulphadiazine and cysteine). The mother solution was heat-treated at 80 °C for 10 min (Moroccan Standard 08.0.125 standard).
- Cetrimide agar was used to isolate *P. aeruginosa*. Afterwards, serological identification was made.
- CHROMagar was used to isolate *Salmonella*. Each isolated bacterium was subsequently identified according to biochemical criteria, using API 20NE, followed by serological confirmation.

The enumeration of major microbial populations in sediment samples was made by the MPN method using appropriate standard media, as mentioned above.

### Disinfection tests

Disinfection tests were performed on the most contaminated samples, which came from reservoir R6. Plastic transparent bottles were aseptically filled with 1 l of the tested reservoir water (R6), transported to the laboratory (at 4 °C, about 2 hours) and directly subjected to the disinfection tests.

### Disinfection by chlorination

The chlorine solution used was in the form of household bleach. To ensure proper disinfection and to specify the minimum quantity of chlorine required for pre-chlorination and water sterilization, the chlorine dose required was

determined by the break-point method. Residual chlorine in the treated water (R6) was measured after 30 min by a colorimetric method. Subsequently, we followed the evolution of the bacterial abundance.

### Disinfection by solar radiation

Disinfection by SODIS is a new approach to improve the quality of water (Sobsey 2002; Rincón Benavides & Pulgarin 2007). Transparent plastic bottles were filled with water from the test site (R6). They were then placed horizontally on a plain black surface. Three of them were exposed to direct sunlight while the other three were protected by aluminium foil (control samples) for about 5 hours. The water was then cooled in a refrigerator before laboratory analysis because during their exposure to solar radiation, their temperature increased to 50 °C after 3 hours.

The effectiveness of disinfection by chlorination and solar radiation was evaluated by studying the abundance of all the studied bacteria before and after each hour of disinfection (total 4 hours of exposure).

### Statistical analysis

The descriptive statistics analyses used to characterize the data set were made by using SPSS (version 10). Principal component analysis (PCA) and analysis of variance (ANOVA II) were conducted and all differences were considered significant at 5%.

## RESULTS AND DISCUSSION

### Evaluation of water quality

#### Physicochemical parameters

The physicochemical parameters of the supplying river waters and the water samples are shown in Table 1 and Table 2, respectively. Overall, the total chemical composition of water was very slightly variable from one site to another (with a gradient upstream–downstream), and also changed over time in the same reservoir.



**Table 2** | Average and standard deviation of physicochemical parameters for the water of the six reservoirs studied during winter and summer periods (2010)

	Temperature (°C)		pH		Conductivity (mg/l)		Chloride (mg/l)		Bicarbonate (mg/l)		Sulphate (mg/l)	
	Winter	Summer	Winter	Summer	Winter	Summer	Winter	Summer	Winter	Summer	Winter	Summer
R1	19.24 ± 0.48	30.49 ± 0.47	8.50 ± 0.06	7.91 ± 0.03	615 ± 10.9	758 ± 8.5	12.83 ± 1.04	19.13 ± 2.20	3.38 ± 0.15	5.01 ± 0.13	26.20 ± 1.11	50.02 ± 8.55
R2	15.22 ± 0.11	26.91 ± 0.12	8.56 ± 0.03	8.20 ± 0.27	9,711 ± 6.9	543 ± 10	17.27 ± 1.11	31.84 ± 3.37	3.61 ± 0.19	6.25 ± 0.11	28.05 ± 2.81	31.09 ± 5.41
R3	16.44 ± 0.18	26.48 ± 0.33	8.74 ± 0.05	7.81 ± 0.07	519 ± 2	906 ± 11	12.00 ± 3.00	21.53 ± 1.75	2.94 ± 0.21	5.02 ± 0.05	32.27 ± 3.56	57.67 ± 5.38
R4	16.05 ± 0.05	26.57 ± 0.51	8.43 ± 0.01	7.76 ± 0.03	529.8 ± 2.3	1,080 ± 30	17.33 ± 2.31	104 ± 7.22	4.03 ± 0.12	6.10 ± 0.15	34.27 ± 4.90	82.47 ± 1.59
R5	15.88 ± 0.39	28.99 ± 0.29	8.32 ± 0.08	7.65 ± 0.04	550 ± 15.5	606.56 ± 7	14.00 ± 2.00	51.8 ± 16.5	2.78 ± 0.28	4.99 ± 0.14	39.31 ± 2.11	47.16 ± 3.10
R6	16.76 ± 0.27	28.2 ± 0.62	8.21 ± 0.02	7.82 ± 0.07	1,440 ± 32	1,204 ± 5.5	13.67 ± 2.08	117.67 ± 3	3.85 ± 0.19	7.12 ± 0.20	40.57 ± 1.50	153.1 ± 5.47
	Calcium (mg/l)		Magnesium (mg/l)		Oxygen (mg/l)		BOD <sub>5</sub> (mg/l)		Oxidability by KMnO <sub>4</sub> (mg/l)			
	Winter	Summer	Winter	Summer	Winter	Summer	Winter	Summer	Winter	Summer		
R1	12.04 ± 1.30	36.94 ± 1.69	225.9 ± 4.9	285.28 ± 10.98	6.74 ± 0.21	6.22 ± 0.28	4.55 ± 0.5	5.35 ± 0.35	1.98 ± 0.37	2.03 ± 0.15		
R2	14.47 ± 0.52	37.08 ± 0.32	236.67 ± 1.3	292.89 ± 5.28	5.81 ± 0.22	5.08 ± 0.42	8.34 ± 1.05	9.14 ± 0.05	3.49 ± 0.08	5.26 ± 0.47		
R3	19.93 ± 0.87	42.09 ± 1.11	222.4 ± 5.16	291.40 ± 2.45	5.07 ± 0.08	3.72 ± 0.63	6.84 ± 0.29	7.38 ± 0.18	3.50 ± 0.14	5.14 ± 0.08		
R4	29.85 ± 1.36	38.04 ± 1.16	267.3 ± 9.3	341.3 ± 3.1	4.92 ± 0.18	3.00 ± 0.46	11.73 ± 0.28	12.28 ± 0.22	3.28 ± 0.29	6.61 ± 0.49		
R5	24.00 ± 0.80	32.00 ± 1.11	213.35 ± 8.3	215.16 ± 6.81	5.19 ± 0.12	4.13 ± 0.48	11.44 ± 0.24	12.99 ± 0.10	3.82 ± 0.07	6.14 ± 0.04		
R6	34.68 ± 1.48	43.93 ± 1.39	302.95 ± 3.2	416.57 ± 10.81	4.73 ± 0.05	2.90 ± 0.35	14.09 ± 0.28	14.55 ± 0.40	4.96 ± 0.09	8.10 ± 0.14		

Comparing the supplying river water and stored water, it seems that there is an increase in the concentrations of the major elements in the stored water, especially for calcium and magnesium, which reached maximum concentrations of 44 and 417 mg/l respectively. This may be related to the calcareous materials used in the construction of the reservoirs and to the geographical position of reservoirs located downstream of the Assif El Mal valley, which is dominated by limestone and dolomite outcrops (Dresch 1941). However, this increase in concentration of minerals does not exceed the safe limits for human consumption according to Moroccan Standards (2002). In all the surveyed tanks, the parameters measuring organic contamination showed significant spatial and temporal fluctuations.

Critical situations were observed during the summer near sites of pollution (R4 and R6). Oxidizability by potassium permanganate is greater than the maximum value permitted in the Moroccan standards (5 mg/l), and BOD<sub>5</sub> is quite high. The average concentration of dissolved oxygen in water reservoirs confirms this organic contamination since it is below the drinking water standards (<5 mg/l), especially in summer. The amount of dissolved oxygen can be reduced

by the activity of bacteria decomposing organic matter (Fekhaoui & Pattee 1993). In contrast, nitrite is present in trace amounts (<0.05 mg/l) and conforms to Moroccan waters standards for human consumption.

All the examined parameters showed relative stability during the rainy period and disturbances occurred when the low water period began.

Concerning the quality of sediments, Figure 2 shows a general upstream–downstream (from R1 to R6) gradient of increasing levels of limestone, total Kjeldhal nitrogen, phosphorus and total organic carbon. The levels of total calcium in the sediment and total hardness (calcium and magnesium) of its supernatant water showed similar trends, confirming the influence of exogenous sources. The endogenous inputs are governed by the sediment–water interactions as a result of the mineralization of organic matter in summer, and by the release of other elements.

### Microbiological quality

In all the reservoirs studied, we observed an increase of bacterial counts in water especially during the summer period.

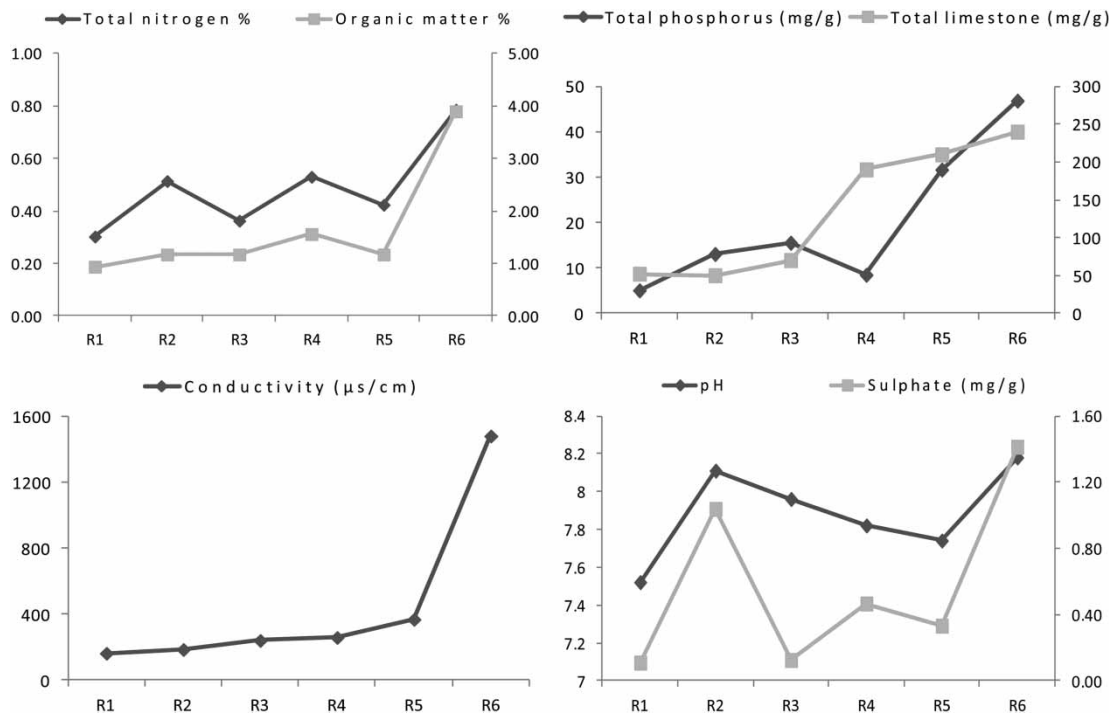


Figure 2 | Values of physicochemical parameters of reservoir sediment samples.

Counts varied from 0.22 to  $30.5 \times 10^3$ , 1.5 to  $24.0 \times 10^3$  and 0.92 to  $8.1 \times 10^5$  CFU  $100^{-1}$  ml in water for FC, EC and TC, respectively (Figure 3(a), (b), (c)). Counts for SD and IE in water ranged from 0.147 to  $7.5 \times 10^3$  and from 2 to  $137.5 \times 10^2$  CFU  $100^{-1}$  ml respectively (Figure 3(d), (e)). Several contemporary studies have shown that the presence of FCs and streptococci in water tanks is a frequent phenomenon. Ahmed *et al.* (1998) observed intense bacterial growth on the inner surface of storage tanks.

An increase in contamination with SRA and *S. aureus* in water was observed especially during summertime and varied respectively from 7.3 to 28.7 and from 3.6 to  $23.83 \times 10^2$  CFU  $100^{-1}$  ml in water (Figure 3(f), (g)). *Salmonella* contamination by different serovars was detected in all reservoirs during the two study periods. Similar contamination was reported by Taylor *et al.* (1999) and Simmons *et al.* (2001), and Khastagir (2008) reported the presence of *Salmonella* in a poorly maintained rainwater reservoir in Rockhampton (Queensland, Australia). Contamination with different serovars of *P. aeruginosa* was also observed, especially in summer.

The various bacterial populations studied changed in the same way with a clear increase in counts during summer. Similar have been reported by other authors (Romney *et al.* 2003; Bou Saab *et al.* 2007).

Regarding the sediment samples, Table 3 shows that they host high densities of faecal bacteria (TC, FC and SD) and SRAs, and pathogenic bacteria: *Salmonella*, *S. aureus* and *P. aeruginosa*. The abundance of faecal bacteria varies significantly from one site to another. Counts for FC, TC and SD ranged between 9.6 and  $440 \times 10^4$ , 8.4 and  $170 \times 10^5$ , 1.3 and  $900 \times 10^5$  CFU/g respectively. Counts for SRA varied between 0.02 and  $520 \times 10^5$  CFU/g and for *S. aureus* between 1.1 and  $9.1 \times 10^3$  CFU/g (Table 3). The abundance of most micro-organisms is greater in the sediment than in the supernatant water, with a significant difference ( $p < 0.05$ ) for all the bacteria studied. This difference is greater for Gram-positive bacteria.

Khastagir (2008) indicated that the sedimentation of a small amount of organic matter may increase the accumulation of nutrients at the bottom of a tank and thus accelerate the growth of bacteria in the tank bottom. Furthermore, this result indicates a greater load of Gram-positive (SD and SRA) in sediment compared to water. Indeed,

previous works indicated that the sediments of shallow waters are home to a rather important bacterial density, because they trap dissolved organic matter from the supernatant water column, especially for SD and SRA (Jørgensen 1983; Mallet *et al.* 2004; Jiang *et al.* 2006).

### Principal component analysis

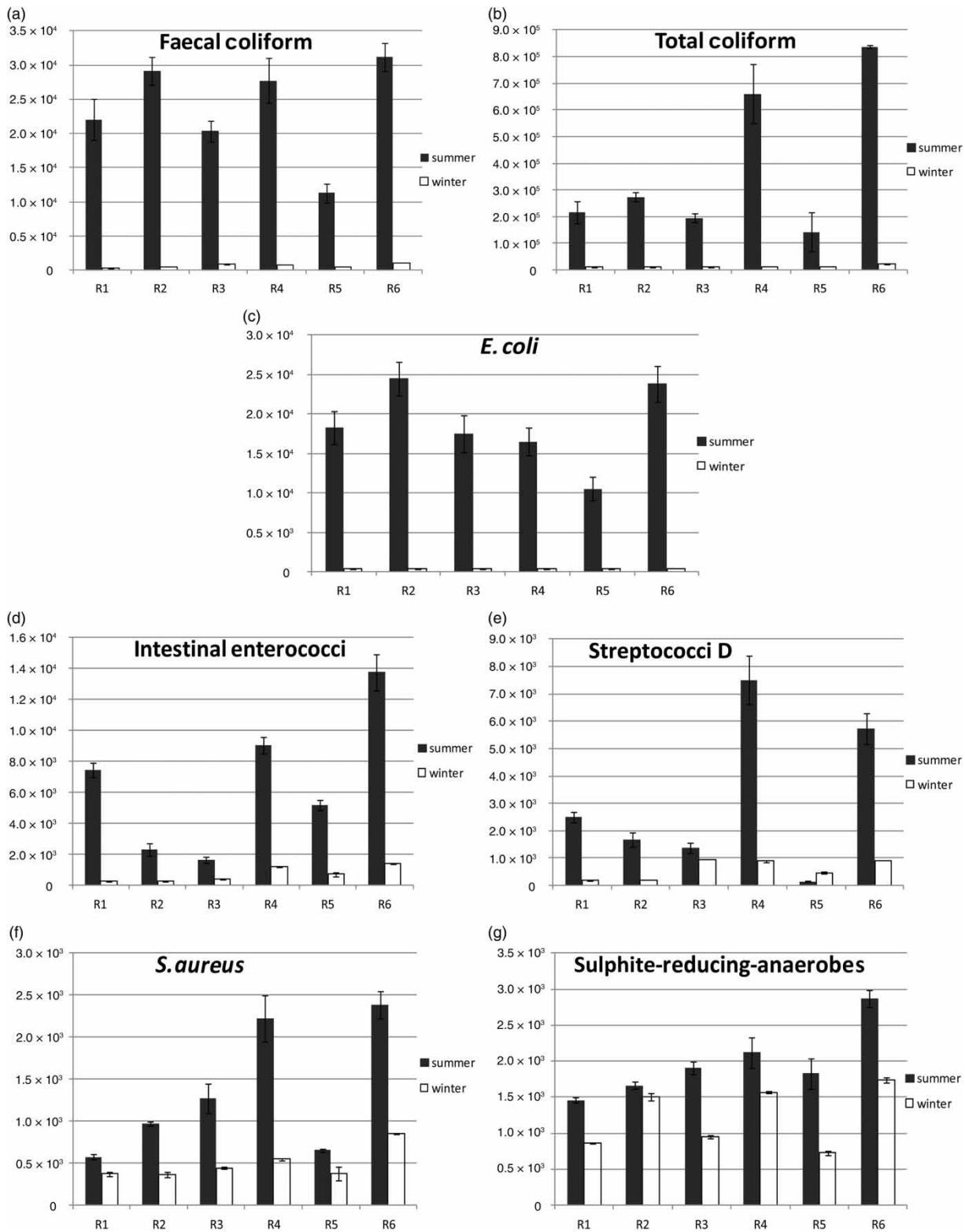
The Eigen values of the two axes F1 and F2 and their contribution to the total inertia are shown in Table 4(a). The coordinates of the variables on the same axes are shown in Table 4(b). These two tables generate a first typological approach for the different variables according to their affinities and their grouping on the first two principal components from their contributions. These determine 80.9% of the total information at a rate of 63.3 and 17.6 of inertia respectively for the factorial axes 1 and 2.

Thus, the projection of these variables on the plan of the two main components ( $1 \times 2$ ), presented on the graph (Figure 4(a)) shows the affinities of the variables from each axis. Figure 4(a) shows that all the variables examined are defined very clearly positive in relation to axis 1. In contrast, pH and dissolved oxygen are negative. This allows us to say that increased concentrations of organic matter are in parallel with those of bacteria, and opposite to oxygen and pH. This distribution is a reason that the organic loading favours the development of micro-organisms whose consumption of dissolved oxygen is followed by acidification of the medium.

A correlation is clearly distinct between Gram-positive on one hand and between Gram-negative on the other hand. In general, the analysis of the correlation matrix between variables shows that all variables studied are strongly interrelated. In addition, we observed a highly significant positive correlation between bacterial load and organic load of sediment on one side, and on the other side between the bacterial load sediment and both the bacterial and organic load of the supernatant water layer.

Figure 4(b) shows a marked seasonal contrast, manifested by a positive correlation in summer and a negative correlation in winter between all sites and axis 1. This is explained by the difference in average loads of all elements analysed: higher in summer than in winter (Table 2 and Figure 3). The water is supplied to the reservoirs only during the winter, which leads to





**Figure 3** | Number of colony-forming units/100 ml in summer (■) and in winter (□). (a) Faecal coliforms; (b) total coliforms; (c) *Escherichia coli*; (d) intestinal enterococci; (e) Streptococci D; (f) *Staphylococcus aureus*; (g) sulphite-reducing anaerobes. Error bars show standard error of the mean.

**Table 3** | The bacterial load in sediments of the reservoirs studied (2010)

	R1	R2	R3	R4	R5	R6
Faecal coliforms (CFU/g)	$9.60 \times 10^4$	$1.80 \times 10^5$	$1.95 \times 10^5$	$9.20 \times 10^5$	$7.20 \times 10^5$	$4.4 \times 10^6$
Total coliforms (CFU/g)	$8.90 \times 10^5$	$1.55 \times 10^6$	$1.68 \times 10^6$	$4.10 \times 10^6$	$8.40 \times 10^{55}$	$1.70 \times 10^7$
Streptococci D (CFU/g)	$5.40 \times 10^5$	$1.34 \times 10^5$	$1.40 \times 10^6$	$7.80 \times 10^6$	$2.40 \times 10^6$	$9.00 \times 10^7$
Sulphite-reducing-anaerobes (CFU/g)	$8.00 \times 10^4$	$1.20 \times 10^5$	$2.00 \times 10^5$	$3.50 \times 10^6$	$8.40 \times 10^4$	$5.20 \times 10^7$
<i>Staphylococcus aureus</i> (CFU/g)	$6.00 \times 10^5$	$1.70 \times 10^5$	$1.10 \times 10^5$	$1.70 \times 10^5$	$3.40 \times 10^5$	$9.10 \times 10^5$
<i>Pseudomonas aeruginosa</i>	+	+	+	-	+	+
<i>Salmonella</i> spp.	+	+	+	+	+	+

(+): present, (-): absent; CFU (colony-forming units).

**Table 4** | (a) Inertia ratio of the first two axes

Axes	Contribution to the total inertia (%)	Cumulative contribution (%)
F1	63.3	63.3
F2	17.6	80.9

**Table 4** | (b) Correlations of variables with axes

Variables	Principal component analysis	
	Axis 1	Axis 2
Faecal coliform	0.72	0.14
<i>Escherichia coli</i>	0.73	0.16
Total coliform	0.71	0.17
Intestinal enterococci	0.71	-0.01
Streptococci D	0.68	-0.02
Sulphite-reducing-anaerobes	0.65	-0.02
<i>Staphylococcus aureus</i>	0.78	-0.06
pH	-0.7	-0.08
Oxygen	-0.65	0.13
Bicarbonate	0.77	0.04
Nitrite	0.39	0.22
Oxidability by $\text{KMnO}_4$	0.67	-0.12
Sulphate	0.68	-0.13
Chloride	0.69	-0.08
Magnesium	0.61	-0.17
Calcium	0.74	0.02
$\text{BOD}_5$	0.41	-0.21

long-term storage of water in other seasons and degradation of water quality.

Moreover, the spacing between the value projections of three sets of samples in summer for each reservoir is explained

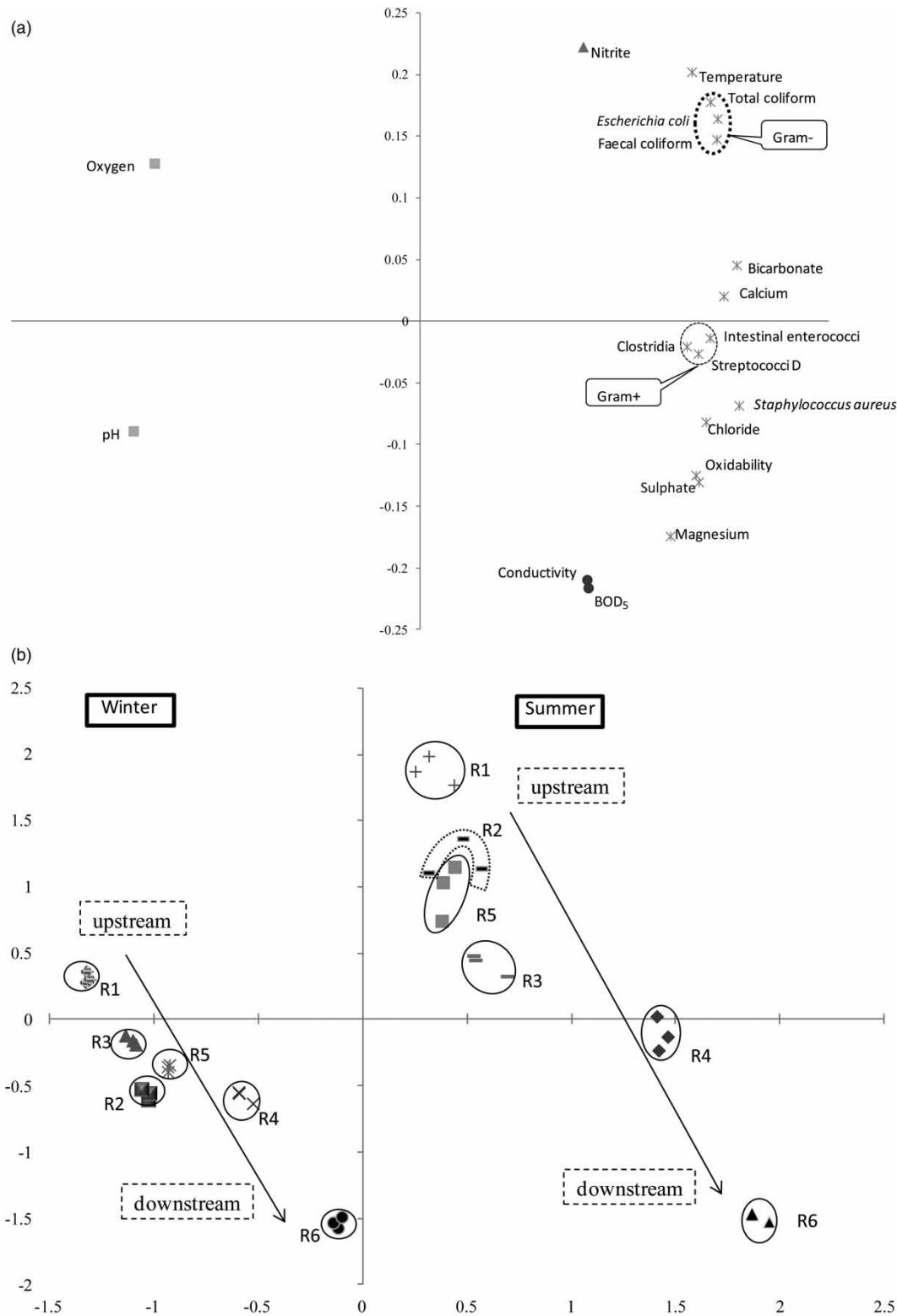
by a fluctuation of the majority parameters analysed, especially bacteriological (large deviation) (Table 4 and Figure 4). In contrast, during winter, these fluctuations disappear and the water quality seems more stable. Figure 4(b) also shows, for the two seasons, a spatial gradient from R1 to R6, indicating an accumulation of chemical and microbiological contaminants from upstream to downstream. This reflects the increase in the impact of pollution caused by the activity of neighbouring populations. Figure 4(b) also shows the isolation of the two reservoirs, among the busiest downstream, R4 and R6 which are situated within or near the largest population centres in the area.

### Disinfection tests

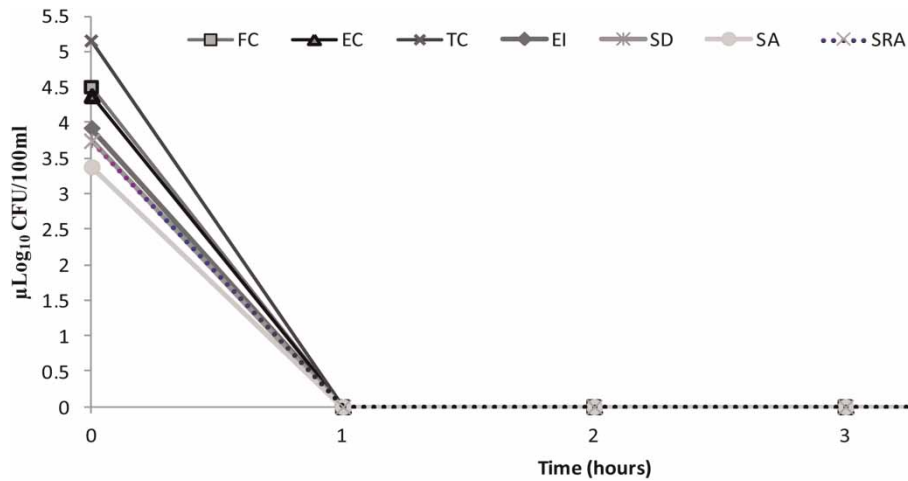
The time evolution of bacterial abundances after injection of chlorine is shown in Figure 5. The results show that chlorination was very effective. Indeed, we noted a maximum reduction of about 5 log units for TC after 1 hour and a total removal of all the studied micro-organisms, even for *Salmonella* and *Pseudomonas*.

Figure 6(a), (b) shows the evolution of bacterial abundances during the exposure of water bottles to solar radiation. The results show that there was an important reduction in bacterial abundance only after 3 hours of exposure to solar radiation. In the exposed bottles, we note an inactivation of 5 log units of TC, 4 log units for FC and 2.7 log units for SF. We also observed inactivation of *Salmonella* and *Pseudomonas* after 2 hours. In contrast, SRA counts were unchanged during the treatment period (5 hours).

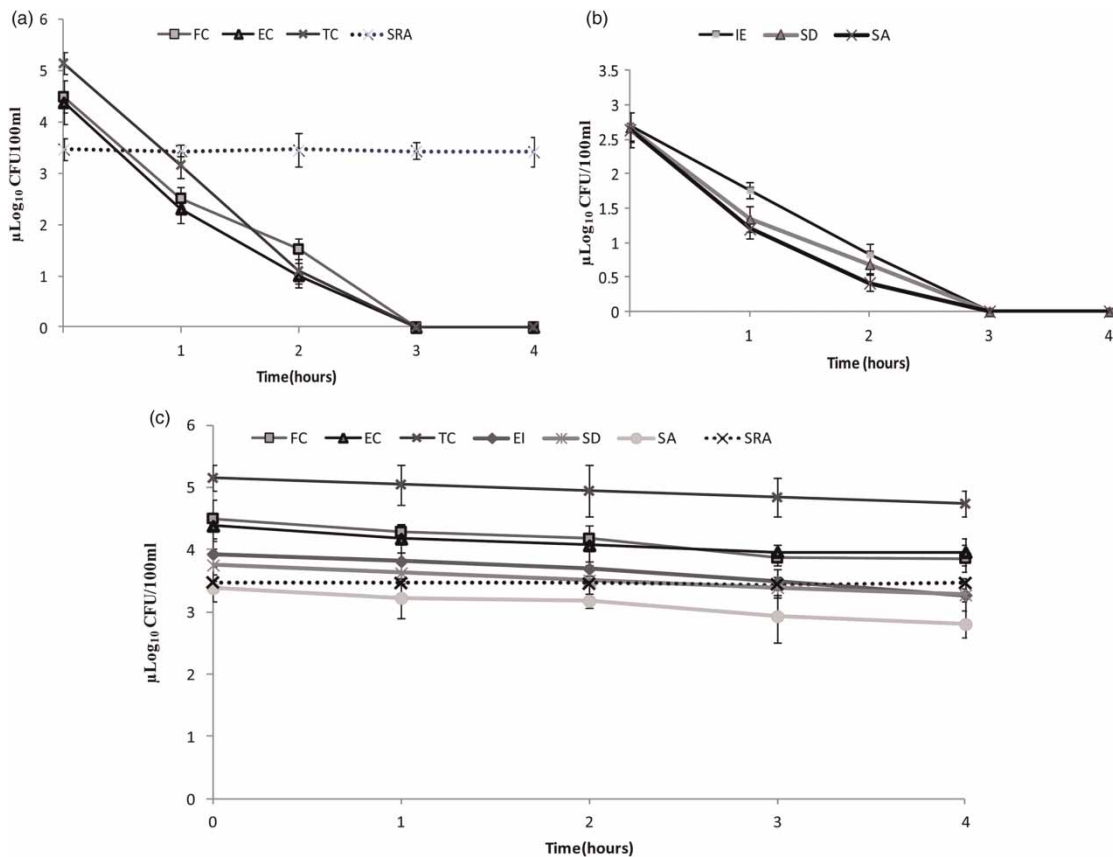
The unexposed bottles (protected by aluminium foil) (Figure 6(c)) have a small inactivation of about 1 log unit



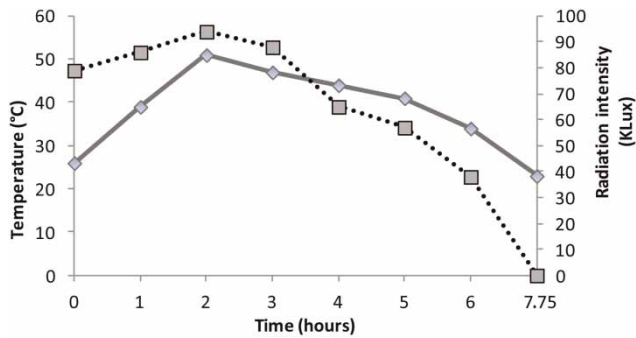
**Figure 4** | (a) Principal component analysis for the physicochemical and bacteriological parameters for all sampling sites during the two study periods. Representation of the distribution of variables; (b) principal component analysis for the physicochemical and bacteriological parameters for all sampling sites during the two study periods. Representation of the distribution for the individual reservoirs.



**Figure 5** | Temporal evolution of bacterial abundances after chlorination (FC: faecal coliforms; EC: *E. coli*; TC: total coliforms; EI: intestinal enterococci; SD: Streptococci D; SA: *S. aureus*; SRA: sulphite-reducing anaerobes).



**Figure 6** | Effect of exposure to solar radiation: time evolution of bacterial abundance in exposed bottles (a) and (b) and (c) unexposed (controls). (FC: faecal coliforms; EC: *Escherichia coli*; TC: total coliforms; IE: intestinal enterococci; SD: Streptococci D; SA: *S. aureus*; SRA: sulphite-reducing anaerobes.) Error bars show standard error of the mean.



**Figure 7** | Effect of exposure to solar radiation: time evolution of temperature (°C) and radiation intensity (kiloLux).

only. Thus, inactivation of the targeted bacteria was significantly higher in the exposed bottles ( $p < 0.05$ ). This results from the absence of transmission of solar radiation in the unexposed bottles, which are subject to the effect of heat only.

By analysing the change in light intensity (Figure 7), we find that the radiation intensity increases to reach, after 2 hours, a maximum of 95,700 lux and then decreases steadily after 4 hours to 300 lux. The temperature changes in the exposed bottles follow those of the radiation intensity and reach 51 °C after 2 hours.

The efficiency of SODIS is due to the synergistic effects of sunlight and heat. This combined effect has already been described by Wegelin *et al.* (1994) and Joyce *et al.* (1996). More than that, SODIS and artificial UVA kill enteric bacteria, most likely by inactivation of the respiratory chain and subsequent exhaustion of ATP (Bosshard *et al.* 2009). Reliability of the SODIS method depends not only on the light dose leading to damage in target cells, but also on possible recovery processes in injured cells after irradiation. So far, no regrowth or recovery of functions in injured bacteria cells has been found (Wegelin *et al.* 1994; Joyce *et al.* 1996; Reed 1997; Oates *et al.* 2005; Berney *et al.* 2006).

By analysing the results of the two disinfection processes, it appears that the chlorine disinfection has a very rapid effect against all micro-organisms tested. However, the presence of organic matter suspended in waters makes the use of this method inadequate, considering the toxicity of organochlorine derivatives that may appear during the chlorination (Wang *et al.* 2007; Legay *et al.* 2011), such as trihalomethanes and halogenated organic carcinogens (IARC 1991; Dunnick & Melnick 1993). This raises acute health

problems (Legay *et al.* 2010), especially in domestic storage tanks (Mahfouz *et al.* 1995). In addition, the presence of a residual chlorine concentration does not always provide real protection and could even hide contamination of the distribution system (Payment 1999).

The effectiveness of SODIS on bacterial load is well established and has already been reported in several studies (Joyce *et al.* 1996; Caslake *et al.* 2004; Boyle *et al.* 2008; Bouziani & Aslah 2009; Mhazo *et al.* 2010). The SRA resisted treatment over 5 hours because of their ability to sporulate; they could easily be reduced by extending the exposure time. Several recent studies have shown that cumulative exposure time (16 hours) to natural sunlight could inactivate 96% of endospores of *Bacillus subtilis* (Boyle *et al.* 2008). In addition, recent studies applying this method of disinfection to highly resistant oocysts of the protozoan pathogen *Cryptosporidium parvum*, showed that exposure for two consecutive days of strong sunshine was generally sufficient for their complete inactivation (McGuigan *et al.* 2006; Mendez-Hermida *et al.* 2007).

According to these results, and taking into account the economic advantages, the SODIS technology also showed the greatest potential to become widely used and is sustainable for improving household water quality to reduce waterborne disease and death in some rural areas.

## CONCLUSIONS

It seems that the temporal evolution of the parameters of water quality in reservoirs is controlled by exogenous inputs in winter (period of reservoirs filling) and endogenous inputs in summer (from the sediment by salting out via the water–sediment interactions during the long period of water storage). Thus, we can say that pollution (mainly organic and microbial) has increased mainly in the R4 reservoir, in the centre of Douar (a small town) and R6, located downstream of Ait Aalouach (the largest city in the valley). In addition, referring to Moroccan standards for safe drinking water, all water reservoirs studied are considered non-potable.

It emerges from this study also that the treatment by chlorination is an efficient method to eliminate bacterial contamination. However, this method seems to have



limitations while there is generation of toxic products because of the presence of organic matter.

The effectiveness of disinfection by SODIS for only 5 hours shows very convincing results. However, its improvement by extending the duration of sunlight exposure could eliminate even SRAs, and thus constitute a more lasting method and the most convenient for water disinfection in rural areas.

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