



Adhesion of *Escherichia Coli* on Fragments of Some Environments Rocks in Aquatic Microcosm: Impact of PH and Biodegradable Organic Compound

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Abstract: A study was conducted in the laboratory to assess the capacity of rocks immersed in water to reduce the abundance of *Escherichia coli* and evaluated the impact of pH and, Biodegradable Organic Compound on adhesion process. These rocks have been chosen according to their representation in the aquatic environment and their chemical composition. The used rocks were collected in four different regions of Cameroon (Central Africa). Rocks used were granite, basalt, micaschist and sandstone. The pH of the medium ranged between 3 and 13 C. U, and Biodegradable organic compound (BOC) concentrations were 0, 2.5, 5, 7.5, 10 and 15 g/l. The duration of the cell adhesion varied from 180 to 1440 min. The highest acidic and basic pH very significantly increases the cells adhesion rate on the substrates ($P < 0.01$). Moreover, when the BOC varies, the average abundances of *E. coli* cells adhered over time ranged from $8.5 \times 10^3 \pm 7.5$ to $57.3 \times 10^3 \pm 11.2$ CFU/cm² on the micaschist, $2.9 \times 10^3 \pm 3.1$ to $81.8 \times 10^3 \pm 14.6$ CFU/cm² on the granite, $3.9 \times 10^3 \pm 5.9$ to $154 \times 10^3 \pm 18.1$ CFU/cm² on the sandstone, and from $3.6 \times 10^3 \pm 5.2$ to $184 \times 10^3 \pm 21.5$ CFU/cm² on the basalt fragments. Therefore, these two parameters should be considered in the methods of treatment of drinking water.

Keywords: *Escherichia Coli*, Cell Adhesion, Rocks, Biodegradable Organic Compound, PH

1. Introduction

Water is very important substance for all living organisms. It represents about 0.6% of the water on the planet, or 8 million km³ distributed in rivers, lakes, reservoirs and groundwater [1]. Freshwater exposure to pollutants is partially due to its greater use for drinking (6%), agriculture (70%), industry (22%), and because of the high demographic explosion that has increased water consumption.

The groundwater recharge proceeds by vertical infiltration of runoff from precipitation, and vertical and horizontal infiltration of surface water [2]. The risk of chemical and biological pollution depends firstly on the characteristics of

the infiltrated water and secondly on the natural properties of the geological strata separating the ground water and the surface. Biological pollutants can be viruses, bacteria, protozoa, yeast, fungi, and arthropods [2].

To solve the problem of water contamination by microorganisms, bacterial adhesion to various substrates has been suggested [3, 4]. Considering the surface water or groundwater, bedrock is mostly composed of rocks of various petrographic and mineralogical natures. In the aquatic environment, the majority of microorganisms prefers a state adhesion on a support (sessile state) rather than free and

isolated in the water (planktonic) [5]. The substrates on which the bacteria adhere in the aquatic environment can be organic or inorganic. Inorganic substrates are generally rocks. Many studies had shown that bacteria adhesion on bedrock varies with respect to the physicochemical characteristics of the aquatic environment, the characteristics of the bedrock and the relevant cell surface parameters such as hydrophobicity, electrostatic charge of the contact surfaces - [5, 6].

Bacteria present in the water and retained by rocky substrates are generally in various shapes. They may be spherical, rod-shaped, comma-shaped, filamentous, etc..., and may possess or not a capsule [7, 8]. Furthermore, these bacteria may have or not surface structures such as flagella or cilia, and be mobile or immobile. The cilia when present, may be polar, peritrichous or mixed [9, 10]. *E. coli* was selected for this study not only because it is the best biological indicator of the quality of drinking water, but also a relevant model organism for the study of the surface colonization [11].

Few studies have so far been conducted on the role of peritrichous cilia in adhesion of microbial contaminants of water on rocky substrates. And, little information is available about variation of organic matter and the pH of medium in adhesion of bacteria to rock fragments. The aim of this study is to assess the importance of pH and biodegradable organic matter on the bacteria adhesion to some rock substrates in aquatic microcosm.

2. Materials and Methods

2.1. Choice of Rock Substrates

The study was focused on four types of rocks: sandstone which is sedimentary rock, granite an acid igneous rock, basalt which is a basic volcanic rock and micaschist a metamorphic rock. These rocks were collected in four different regions of Cameroon (Central Africa). These rocks have been chosen according to their representation in the aquatic environment and their chemical composition. These rocks were subjected to mineralogical analyzes by X-ray diffraction and to X-ray fluorescence geochemical analyzes.

The rock used contain minerals such as quartz, gibbsite, olivine, clinopyroxene, plagioclase, opaque minerals, goethite, sericite, zircon, and traces of kaolinite. These minerals are present in different proportions and may be absent depending on the rock in question. These rocks are also composed of silica, aluminum, iron, manganese, calcium, potassium, magnesium and phosphorus. The proportions of silica, for example, represent 69.21%, 43.91%, 75.35% and 68.32% by weight of oxide for sandstone, basalt, granite and micachist respectively [3].

Four fragments A, B, C and D of rectangular shaped (0.9 cm wide, 3.24 cm length and 0.9cm in height) of the same surface structure were obtained from each type of rock in triplicate. The total surface of each fragment was 13.28 cm².

2.2. Identification of *E. Coli*

According to the protocol described by Tamsa Arfao [12], the identification of *Escherichia coli* has been done on Endo agar (Bio-Rad), incubated at 44°C for 24 to 48 hours. After subculture of red colonies with a metallic sheen on Plate Count Agar, the following biochemical tests were carried out: the research for oxidases, catalase and nitrate reductase enzymes, degradation by fermentation of glucose and lactose, mobility, production of gas and of hydrogen sulfide.

2.3. Preparation of Bacterial Suspensions

The kinetics of bacterial adhesion were evaluated using bacteria suspensions. However, the presence of exopolysaccharides, excreted by the bacteria during the culture phase, may interfere with the optical density measurements. It appeared necessary to optimize the method of preparation of bacterial suspensions in order to overcome this problem. Experimental protocols proposed by Rubio [6] were used.

Isolated *E. coli* cells were preserved in cryotubes (Marine Broth + 15% glycerol) at -80°C and -20°C and then regenerated after thawing, in 9 ml of Marine Broth (Difco Laboratories).

Bacterial suspensions were obtained by incubation for 24 hours. A vials containing 99 ml of Marine Broth were inoculated with 1 ml of bacterial suspension. Bacterial cultures were then placed under stirring at 250 rev/min at 25 °C for 24 hours. Bacterial cultures were filtered through GFC filter (Whatman) to separate the flocks of *E. coli*. The cultures were centrifuged (Sigma 3K15, rotor 12 150-H) at 5000 g for 10 min. at 25 °C. The pellets of *E. coli* were resuspended in NaCl solution (0.85%).

A second centrifugation was performed to "wash" the bacteria. The protocol of Pembrey et al [13] recommends a centrifugation for 10 min at 5000 g and 25°C (protocol 1), while the protocol of Zhang *et al.* [14] recommends a centrifugation for 30 min at 11300 g and 25°C (protocol 2). These two protocols were used. Bacterial pellets were resuspended in a NaCl solution (0.85%). The optical density of the bacterial suspension was adjusted to 0.8 at 400 nm, which corresponded approximately to 2 x 10⁸ CFU/ml.

The suspensions of *E. coli* used in the adhesion experiments were performed in artificial sterile sea water (washing and final suspension) to be in conditions of the aquatic environment [6].

2.4. Impact of pH

The pH of these solutions was adjusted to 3, 5, 7, 9, 11 and 13 using NaOH or HCl 0.01M, and then the solutions were sterilized in an autoclave for 30 minutes at 120°C before use. After sterilization, pH of all solutions were checked again to make sure they were not changed.

2.5. Impact of Biodegradable Organic Compound

The biodegradable organic matter used was peptone. It is a protein containing 18 amino acids in different proportions. It is mainly composed of glutamic acid (17.5%), leucine (7.5%) and histidine (6.5%). Tryptophan and glycine represent only 1.5% and 1.75% respectively. The BOC concentrations were 0 g/l, 2.5 g/l, 5 g/l, 7.5 g/l and 10 g/l. These concentrations were selected to mime the concentrations of organic matter found in wastewater in Cameroon [15].

2.6. Experimental Protocol

The adsorption tests were carried out by immersing the rock fragments (samples suspended with 0.001 mm diameter wire) in solutions by varying the pH on one hand and biodegradable organic matter on the other hand. After 180-1440 minutes incubation, rock substrates were rinsed twice by immersion in physiological saline (NaCl: 0.85%). After drying in a sterile environment created by the Bunsen burner flame, the substrates were introduced successively into three test tubes containing 10 ml of physiological sterile saline (NaCl: 0.85%). Unhooking of bacterial cells attached to different rock fragments was performed by vortex agitation of substrates in 10 ml of sterile physiological saline using a TFA-9 tube agitator of BUNSEN brand operated at increasing speeds from 30 to 50 rpm for about 20 seconds. The increase of unhooking speed in fact leads to obtain the maximum unhooked cells [12, 16]. The unhooking of cells was well done for each rocky fragments removed from each flask, three times, to maximize the detachment of *E. coli* cells adhered. The three suspensions from the dropout were mixed and the total volume of the bacterial suspension obtained was 30 ml. This suspension was used to determine the number of cells adhered to each substrate. The cell counts were made in the selective medium (Endo). The results were expressed as colony forming units per milliliter of water (CFU/ml) of coming from unhooking suspension and reported to centimeter square of rock fragment (CFU/cm²) [4]. The data were transformed into logarithmic values to enable an easy interpretation. For each experiment, the analysis was done in triplicate. The differences were not considered because the curves were close [17].

2.7. Data Analysis

Speed adhesion of bacteria on rock substrate were assessed by determining the Excel logarithmic regression line of bacterial cells adhered to each three-hour incubation period. The slope ratio of the line over three hours gives the adhesion rate of the bacterial cells per hour. The results were expressed as cells adhered/cm²/hour [12]. These speeds allow us to appreciate the presence of *E. coli* peritrichous cilia on the adhesion process of this bacterium to the rocky substrates in the presence or absence of BOC with respect to the pH of the medium.

The relationship between the abundance of bacterial cells adhered and concentrations of BOC on one hand and between the abundances of bacterial cells adhered and pH of solution on the other hand, were assessed by the Spearman correlation test for each duration cell adhesion. Comparisons of data were performed using the test H Kruskal -Wallis.

3. Results and Discussion

3.1. Effect of pH Medium and Adhesion Speeds with Respect to the pH of the Solution

The abundances of *E. coli* cells adhered per unit surface of the substrate have varied with respect to the pH of the medium and different incubation periods. The analysis revealed that when the pH of the solution is very acidic (pH=3), *E. coli* cells adhered on substrate seem to be very low. When the pH of the solution ranged from 5 to 11 CU, the *E. coli* cells seem to move better, which would promote the adhesion of these cells on rock fragments. However, at pH 13, the adhesion of *E. coli* become rare after an incubation period of 540 min on the micaschist, granite and basalt fragments. The same results were observed on sandstone after 180 min of incubation (Figure 1). It has been noted that the mean abundance of *E. coli* cells adhered ranged from $0.26 \times 10^2 \pm 0.4$ to $36.5 \times 10^2 \pm 25.1$ CFU/cm², $0.03 \times 10^2 \pm 0.04$ to $21.1 \times 10^2 \pm 17.3$ CFU/cm², $0.1 \times 10^2 \pm 0.1$ to $37.3 \times 10^2 \pm 32.6$ CFU/cm², 0 to $35.8 \times 10^2 \pm 31.3$ CFU/cm² respectively in the solution with pH 3, 5, 7, 9, 11 and 13 C. U (Table 1).

Table 1. Average abundances (and standard deviation) over incubation time of adhered cells to rock substrates at each pH considered.

Adsorbent considered	Values of pH considered and average abundances ($\times 10^2$) (Standard deviation)					
	3	5	7	9	11	13
Micaschist	0.26 (0.4)	36.5 (25.1)	25.1 (30.3)	12.4 (16.53)	1.51 (1.12)	37.95 (32.43)
Granite	0.03 (0.04)	19.7 (22.3)	21.1 (17.3)	4.01 (2.23)	0.53 (0.17)	89.04 (161.65)
Sandstone	0.10 (0.1)	37.3 (32.6)	25.7 (22.2)	16.8 (20.5)	1.23 (0.96)	147.03 (271.67)
Basalt	0 (0)	35.8 (31.3)	31.6 (35.9)	19.3 (28.4)	2.55 (1.97)	85.28 (127.27)

The Figure 2A depicts the hourly adhesion speeds of *E. coli* with respect to rocks sorbents. The adhesion speeds of *E. coli* cells, very low at pH = 3, 9 and 11, increased at pH = 5 and 7 with the highest values of 1.64×10^2 cells/cm²/hr. In

general when the pH of a solution ranges, two trends of *E. coli* adhesion were observed. The tendency 1 showed that when the pH fluctuated between 3 and 7 C. U., *E. coli* adhered more when the pH of solution was more acidic. The

trend 2 was obtained by varying the pH 9 and 13. *E. coli* adhesion was faster when the pH was too basic (pH = 13).

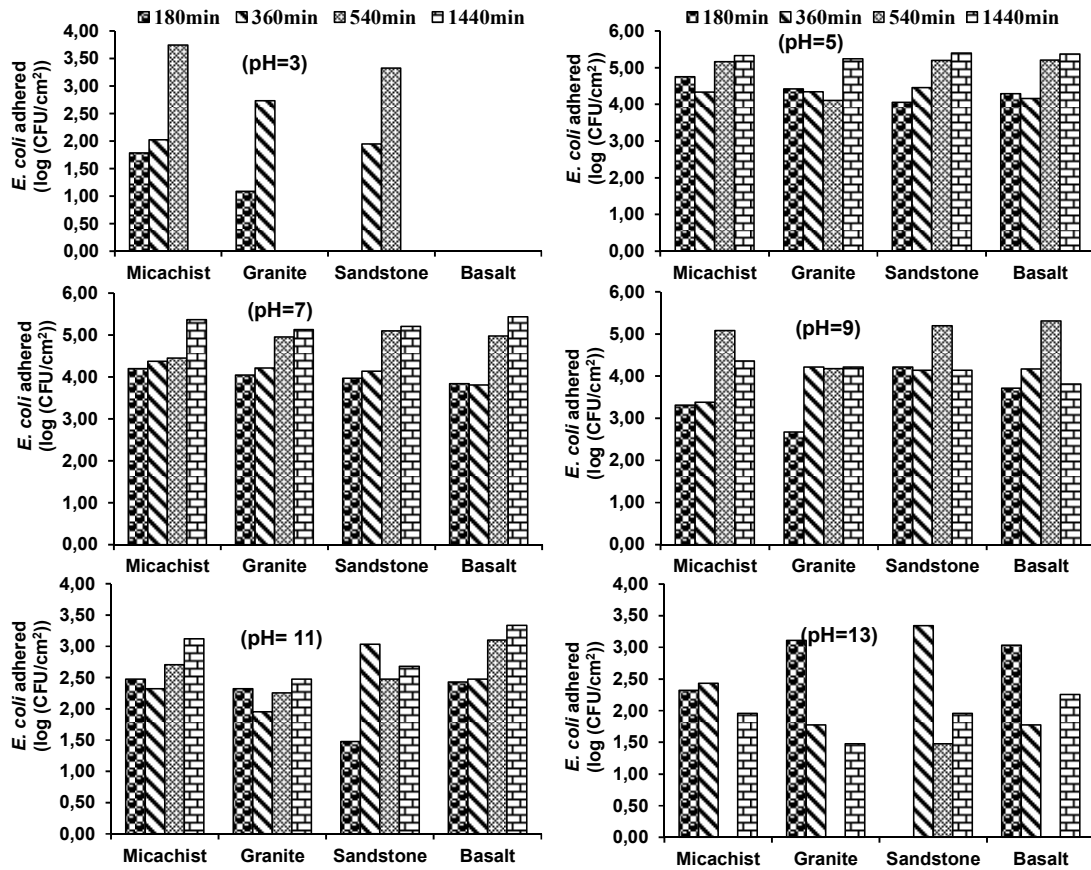


Figure 1. Variation of the means of abundance of *E. coli* cells adhered with respect to rock species at each pH considered.

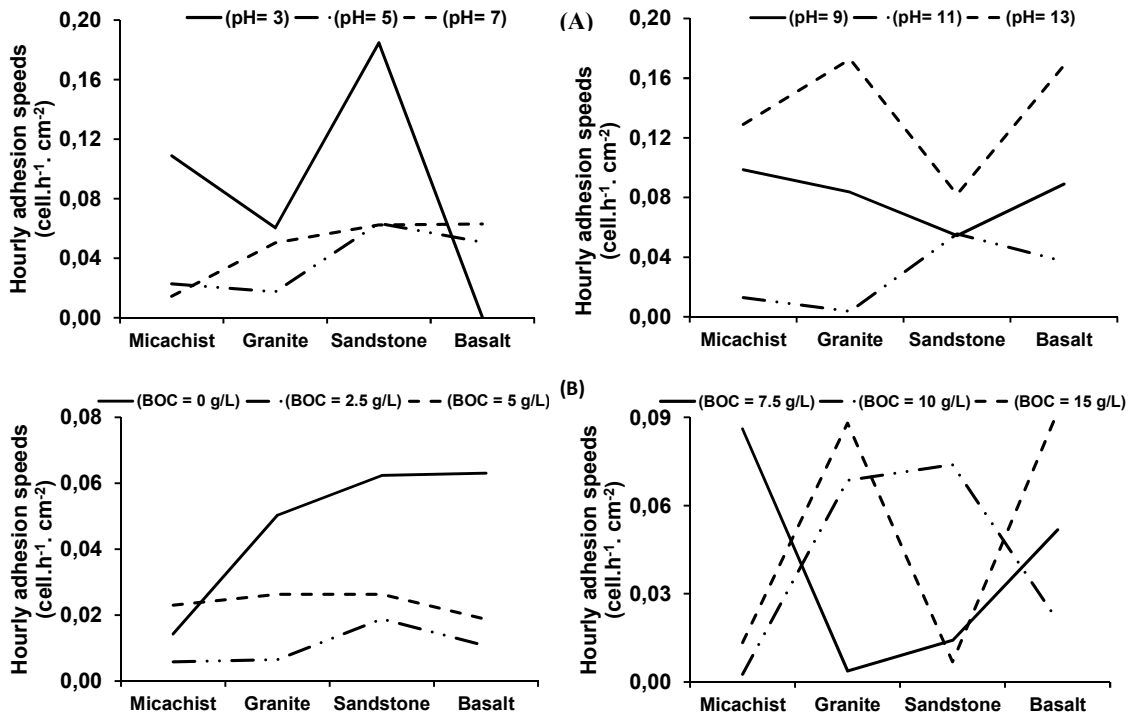


Figure 2. Hourly adhesion speeds of *E. coli* with respect to rocks sorbents, pH (A) and BOC concentrations (B).

3.2. Effect of the Presence of BOC, and Adhesion Speeds with Respect to the BOC Present in the Solution

Generally, cell adhesion of *E. coli* to bedrock varied with respect to the concentrations of biodegradable organic compound present in solution, the incubation period and

substrate considered (Figure 3). Indeed, it was observed that the presence of organic matter in aquatic microcosm promoted the adhesion of *E. coli* cells to bedrock. The organic matter would induce the proliferation of cells previously adhered to rocks.

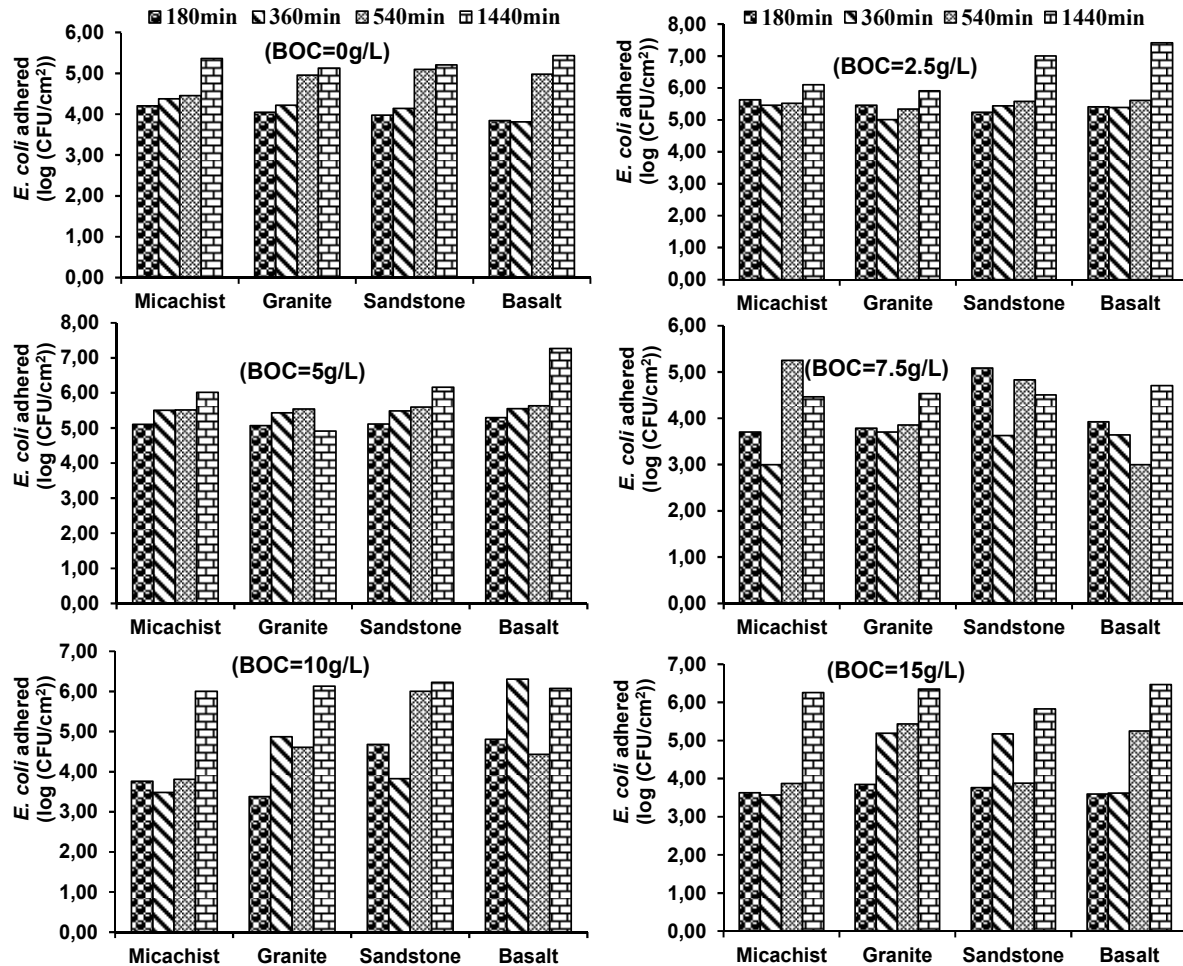


Figure 3. Variation of the means of abundance of *E. coli* cells adhered with respect to rock species and concentration of biodegradable organic compound in the medium.

Table 2 shows the mean abundances of *E. coli* cells adhered to different rock fragments. It appears that the mean abundances of *E. coli* cells adhered with respect to the incubation periods. The average abundances of *E. coli* cells adhered over time ranged from $8.5 \times 10^3 \pm 7.5$ to $57.3 \times 10^3 \pm$

11.2 CFU/cm^2 on the micaschist, $2.9 \times 10^3 \pm 3.1$ to $81.8 \times 10^3 \pm 14.6 \text{ CFU/cm}^2$ on the granite, $3.9 \times 10^3 \pm 5.9$ to $154 \times 10^3 \pm 18.1 \text{ CFU/cm}^2$ on the sandstone, and from $3.6 \times 10^3 \pm 5.2$ to $184 \times 10^3 \pm 21.5 \text{ CFU/cm}^2$ on basalt.

Table 2. Average abundances (and standard deviation) over incubation time of adhered cells to rock substrates in the presence of biodegradable organic matter.

Adsorbent considered	Biodegradable concentration and average abundances ($\times 10^3$) (Standard deviation)					
	0 g/l	2.5 g/l	5 g/l	7.5 g/l	10 g/l	15 g/l
Micaschist	25.1 (30.3)	10.6 (8.5)	8.5 (7.5)	12 (8.7)	57.3 (11.2)	8.5 (16.9)
Granite	21.1 (17.3)	6.6 (5.8)	3.8 (2.3)	2.9 (3.1)	81.8 (14.6)	12.3 (19.3)
Sandstone	25.7 (22.2)	50.9 (9.1)	10.6 (11.1)	12.6 (11.3)	154 (18.1)	3.9 (5.9)
Basalt	31.6 (35.9)	125 (23.8)	88.3 (16.9)	3.6 (5.2)	184 (21.5)	14.4 (26.5)

It was observed that in the presence of BOC, adhesion speeds become arbitrary. Figure 2B shows the variation of adhesion speeds of *E. coli* on rock substrates in the presence of BOC. It appears that in the presence of BOC at the concentration of 7.5 g/l, *E. coli* cells adhered on granite, sandstone and basalt with adhesion speed of 1.08×10^2 cells/cm²/h and the micaschist with adhesion speed of 22 cells/cm²/h. With the exception of granite, the *E. coli* adhesion speeds in the presence of other concentrations of BOC were higher. Indeed, in the presence of BOC, *E. coli* found no interest to adhere on rocky substrates.

3.3. Correlations Between Considered Parameters

The correlation coefficients "r" Spearman between bacterial abundance and the incubation periods for each

Table 3. Correlation between cells abundances of *E. coli* and during incubation period with respect to the pH and biodegradable organic compound for each rock used.

Abiotic parameters and rock used	pH				BOC			
	Micaschist	Granite	Sandstone	Basalt	Micaschist	Granite	sandstone	Basalt
Incubation duration	0.178	0.027	0.232	0.151	0.560**	0.490*	0.538**	0.506*

ddl: 23 **: Most Significant correlation (p<0.01), *: Significant correlation (p<0.05)

3.4. Comparisons Between the Considered Parameters

The mean abundances of bacteria adhered on rock fragments were compared with each other using the H test Kruskal-Wallis. The different P-values were presented in Table 4 and 5. It appears that significant differences were observed between the mean abundances of *E. coli* cells adhered to all bedrock when the pH of aquatic microcosm varies (p <0.05). In the contrary, in the presence of BOC no significant difference were obtained (Table 4). When considering the variation of the incubation period, the rock substrates and abiotic factors of the environment, a

rocky substrates considered were evaluated and presented in Table 3. Moreover, there were no significant correlation between the incubation period and the abundance of cells adhered when the pH of solution varies. In the presence of BOC, it was observed a very significant and positive correlation between incubation period and the abundances of *E. coli* adhered on mica (r = 0.560; p <0.01). The similar result was obtained on sandstone (r = 0.538; p <0.01). Moreover, significant and positive correlations were observed in granite and basalt (Table 3). Thus, the adhesion of *E. coli* cells increase with increasing of incubation periods and concentrations of BOC present in aquatic microcosm. However, no significant correlation was obtained between bacterial abundance and abiotic factors (pH, BOC) of the aquatic microcosm.

significant difference between the adhered cells abundances on the micaschist was observed when the BOC vary in aquatic microcosm (Table 5).

Table 4. Comparison between cells abundances of *E. coli* adhered on each rock fragment when pH and BOC vary in aquatic microcosm.

Abiotic parameters considered	Rock used and risk values of probability			
	Micaschist	Granite	sandstone	Basalt
pH	0,003*	0,003*	0,003*	0,001*
BOC	0,024	0,053	0,076	0,021

ddl: 23 *: différence significative (p<0.05)

Table 5. Risk values of probability related to the comparison between the abundance of *E. coli* adhered on each rock fragment and duration of cell adhesion in each experimental condition.

Abiotic parameters considered and duration of cell adhesion process	Rock used and risk values of probability			
	Micaschist	Granite	Sandstone	Basalt
Duration of cell adhesion (pH)	0.762	0.885	0.541	0.888
Duration of cell adhesion (BOC)	0.026*	0.137	0.065	0.066

ddl: 23 *: différence significative (p<0.05)

The results revealed that *E. coli* adhered on rock fragments at different degrees. This adhesion varies with respect to the incubation periods, the rock substrate, the pH and the presence of biodegradable organic matter in aquatic microcosm. The abundance of bacteria adhered significantly vary with respect to the environmental conditions. Indeed, studies on the adhesion of microorganisms on solid substrates revealed that adhesion is a process that unfolds reversibly and irreversibly [18]. Exopolymers synthesis, starting from the early stages of adhesion continues for the maturation of biofilm [19] and the matrix can then take up to 75-95% of the volume of a mature biofilm [20].

The adhesion of bacterial cells on the bedrock with respect

to incubation periods was significantly influenced by the pH of the aquatic microcosm. This influence was significant when the pH was neutral. This result was corroborated with that of Gordon *et al* [21]. Other studies revealed that most bacteria grow better at the pH near neutrality or slightly alkalinity. Balebona *et al* [22]. have also observed that *Sparus aurata* adheres better to the glass at neutral and basic pH than acidic pH. Stanley [23] showed that the attachment of the bacteria may vary depending on the pH and was generally high at the metabolic optimum. Indeed, the pH of the variation environment, the surface charge of the microorganisms as well as solid support followed the displacement of the equilibrium ionization

(protonation/deprotonation) of functional groups exposed according to their pKa which may result in a reduction or increase in favorable electrostatic interactions or unfavorable repulsive adhesion of microorganisms [24, 25]. Moreover, it was suggested that when the pH of solution is unfavorable (very acidic or very basic) bacteria can adopt behaviors that facilitate their adhesion on substrates [24].

This study also revealed that *E. coli* adhered more when the pH of the solution was more acidic or more basic. This could be explained by the fact that the highly acidic and highly basic pH would be a limiting factor on the movement of *E. coli*. Thus, the bacteria develops a survival model which is the acceleration of the movement with the peritrichous ciliature of *E. coli* to bedrock. The *E. coli* cell into the activity of flagellin molecules in its peritrichous ciliature which promotes its rapid progress toward the substrate for adhesion and biofilm formation [26]. In other, the colonization of the surface of the rocky substrate by cells during the incubation periods, is concomitant when increasing the concentrations of BOC. It was also noticed that bacterial adhesion to substrates was limited when environmental conditions become unfavorable: limiting the availability of oxygen, decreased concentration or modification in the nature of available nutrients [27]. Under these conditions, the bacteria that were attached may separated themselves from the support to find a favorable environment for their development [28]. However, when organic matter is present in the medium, it may be a coating on the surface of substrates and a modification of their surface properties, which would promote the hydrophobicity and cell adhesion [6]. In addition, once the bacteria was adhered to the surface, its multiplication leads to the formation of colonies that will cover all or part of the surface according to the surface properties of bacteria and materials and result in the formation of biofilms. The structure of the biofilm depends on environmental conditions such as the carbon source or hydrodynamic regime [23].

In the presence of BOC and when the pH of solution varies, *E. coli* cells adhere on surfaces of micaschist, granite, sandstone and basalt. Some authors have however revealed that adhesion was influenced by the mineralogical properties of certain rocks [3]. And among the effects of rock substrates on the adhesion of microorganisms, they are chemical elements which play the role of nutrients or electron acceptors [29]. The bacteria are negatively charged, they can easily attach to a surface also negatively charged such as quartz or feldspath in the conditions of surface waters. Grain sediment of quartz or feldspath are often covered with a crust of clay and iron oxyhydroxides, promoting trapping bacteria [30, 31]. Scholl *et al* [30] have shown that bacteria are much more present on limestone, iron oxyhydroxide on quartz or muscovite positively charged at a pH close to 7. These results are similar to those of Roberts [31] who observed that after 12 months, in groundwater, only the magnetite inclusions showed significant bacterial colonization and that the presence of aluminum was an unfavorable factor for bacterial growth either on a silicate or glass support ($Al_2O_3=20\%$).

This inhibition would be significant from Al_2O_3 contents as low as 1.2%.

The comparison tests show significant differences between the mean abundance of each species when varying the pH and BOC concentrations in aquatic microcosm. The differences observed between the mean abundance of *E. coli* cells adhered were linked to environmental stress caused by the variation of pH of the solution and the carbon source. Stanley [23] believed that fixing bacteria of the same species can vary depending on the pH and is generally higher in the metabolic optimum (optimum pH and carbon source). The differences between the mean abundances of *E. coli* adhered to substrates, mobile bacteria with peritrichous flagella were also observed ($P<0.05$) between the periods of incubation. They would be the resultant of the physicochemical properties of this bacterial species related to the presence of surface appendices and properties of the bacterial wall [9, 32]. The wall of Gram negative bacteria such as *E. coli* does not consist of teichoic and lipoteichoic acids. In adhesion process, these acids can intervene on the physicochemical interactions between a given medium and bacteria. In the aquatic environment, for the same bacterial strain, these characters were fluctuated according to the physicochemical properties of the solution [32].

When considering *E. coli* speed cell adhesion, it was observed that *E. coli* adhered more quickly when the medium is very acidic or very basic and this speed becomes ordinary in the presence of biodegradable organic compound. This result could be explained by the presence of the peritrichous cilia on *E. coli* cells. The ability to adhere on variable surfaces is influenced by specific surface proteins and cell appendix [33]. Bacterial appendices were often useful in cell adhesion, although they are not essential for the remaining cells on a solid surface. Type 1 pili and flagella have been identified as essential for the initial attachment of bacteria on a solid surface, and type 4 pili were important in the movement of bacteria on an adsorbent [34]. It was noted that the abundance of these appendices is irrespective of the temperature of the environment [35]. In addition, the ability of bacteria to "swim" with the flagella by propulsion is important for the initial attachment. Unlike the flagellar swimming, cell motility by contractions only occurs when cells are attached to a surface and the bacteria themselves can slide on this surface [36].

However, Belas & Colwell [37] revealed that the adsorption of *Vibrio parahaemolyticus* follows the Langmuir adsorption isotherm which is an adsorption-type called kinetic surface saturation when conditions are favorable for the production of lateral flagella. When conditions were not favorable for the production of lateral flagella, bacterial adsorption did not follow the Langmuir adsorption isotherm, but rather proportional adsorption kinetics was observed. The adsorption of certain bacteria to polar flagellation have presented surface saturation kinetics. However, the bond index (number of binding sites time affinity of bacteria on the surface) of polar flagellation bacteria differed substantially from that of bacteria flagellated laterally. This observation

showed that polar flagella in bacteria can adsorb to the surface by a mechanism different from that used by flagellated bacteria laterally. Meanwhile, Lapidus *et al.* [38] had reported that when the bacterial cells were attached to a surface by their flagella, many of them are moving, and they thought that the engine that drives the flagellum is divided in two states, existing either in the clockwise direction or the anticlockwise direction. They also showed that a third state of flagella movement is that of the pause, the duration and the beat frequency that are affected by chemotactic stimuli. They also noted that the *E. coli* and *Salmonella typhimurium* cells are wild type cells whose flagella are intermittently in rest.

4. Conclusion

Adhesion of *E. coli* on bedrock in the aquatic environment is influenced by the pH quite acidic or basic. These extreme pH values would cause the acceleration of the migratory movement to aquatic rocky fragments. This migratory movement constitutes survival mean of facing hard environmental conditions. Biodegradable organic matter significantly increases the adhesion of *E. coli* cells and the culturability of *E. coli* initially adhered. These two parameters should be taken into account in the treatment methods of drinking water.

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