

Algerian Sahara's Chott: Study and Characterization

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Abstract:

Sebkhas, chotts, saline soils, etc. represent the hypersaline environments, and are typical examples of severe environments harboring a very intriguing microbial flora. In this regard, this paper aims to characterize the bacteria in the waters of the chott of An El-Beida in the region of Ouargla (South of Algeria). The pH of the water samples tested ranges from 6.6 to 8.39, the water is neutral to alkaline, the temperature ranges from 8 to 10 degrees Celsius, and the electrical conductivity ranges from 2.4 to 6 ds/m, showing extremely saline water. These qualities allowed us to choose a microflora that is particular to this environment. The microbiological analysis of these samples identified fifteen isolated strains, with bacterial densities ranging from $0.27 \cdot 10^4$ to $2.236 \cdot 10^4$, including the species *Escherichia coli* 2, *Flavobacterium meningosepticum*, *Shigella* sp., and *Aeromonas salmonicida*. This may help to explain the richness of water in this ecosystem by a variety of significant microflora. Our research on NaCl tolerance has shown that some isolated strains of euryhaline bacteria can withstand a NaCl concentration of up to 35%. The strain (S8) demonstrated a substantial antifungal capacity in the antagonist test, which can be attributed to the fact that it is highly competitive with *Aspergillus carbonarius* and *Penicillium* sp. This particular extreme ecosystem's microbial research was effective since we were able to identify a sizable amount of specific microbial diversity.

Keywords: Waters, Ecosystem, Characterization, Bacteria, Sahara of Algeria.

Introduction

All aquatic environments have a large abundance of microorganisms. The majority of these bacteria grow in environments with good water supply, temperatures between 20 and 30 °C, air pressure, and neutral pH values [1]. Other microorganisms that are classified as extremophiles, however, can inhabit settings that are extremely harsh in terms of temperature, pH, salinity, depth, or other physico-chemical factors. These environments are distinguished by physico-chemical factors that are thought to be hostile to the existence of the majority of species. Many techniques have been established since the 1980s for identifying and figuring out the makeup of halophilic microbial communities in hypersaline environments. Understanding the relationships between the microbial communities and how this sort of ecosystem functions depends on the study of these communities [2, 3]. There are various hypersaline environments in Algeria, some of which are wetlands and are designated as Ramsar areas. Multiple research projects have shown the richness of the fauna and flora in these habitats. However, there hasn't been much research done on how to analyze these environments from the perspective of microbial diversity. We carried out a study to characterize certain halophilic bacteria from the waters of the chott of Ain El Beida, in the Sahara of Algeria, in light of current knowledge. Therefore, during this work, we made an effort to identify the halophilic bacteria present in this environment. Water samples were taken from chott (outlet of untreated water) and subjected to physico-chemical analyses in order to characterize various bacteria in an aquatic ecosystem.

Presentation of the chott Aïn El-Beida

The chott of Aïn El-Beida constitutes the low point of the city of Ouargla. Lying in a North-West, South-East direction over a length of 5.3 km, its width varies from 1 to 1.5 km, not exceeding 1.50 m in depth [4]. The water supply of the chott comes from the water table, the level of which varies according to the season and the actions of man (drainage of the palm grove and irrigation).

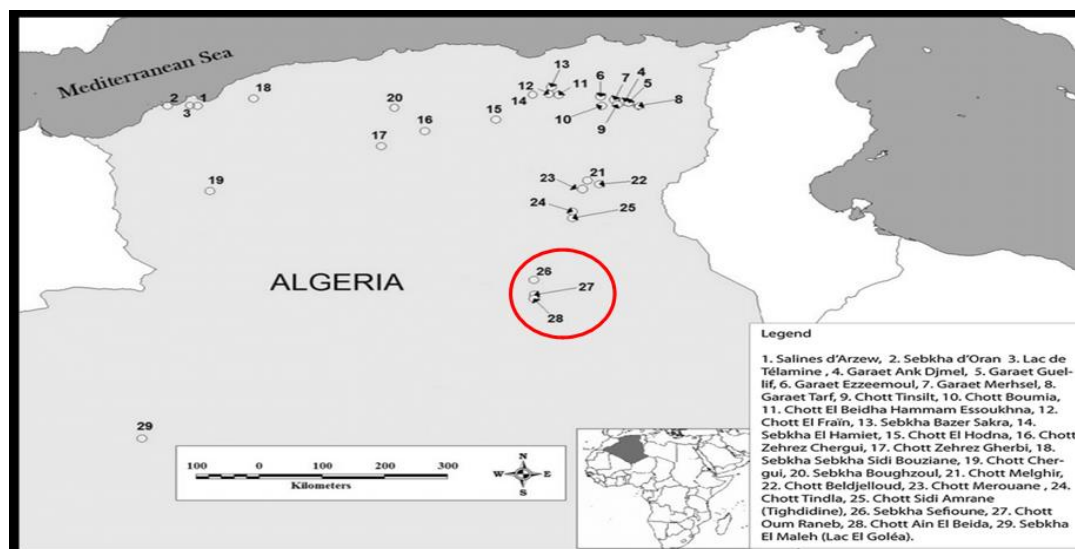


Fig. 1 Geographical location of chotts and sebkhas in Algeria (non-exhaustive list) [5]

- Material and Methods

Before choosing the sampling station, a visual recognition such as the color of the water, the direction of the wind or any other relevant observations (bad odours, etc.). Water samples from the chott are taken at three different points and from a depth ranging from 1cm to 50cm.



Fig. 2 Sampling Site

2.2. Measurement of physicochemical parameters of samples

Measurement of the physico-chemical parameters of the samples: pH, temperature and electrical conductivity are measured in situ. The pH in the aquatic environment is essentially a function of the chemical composition of the aqueous solution and the activity of the organisms [6]. Electrical conductivity makes it possible to assess the quantity of salts dissolved in water [7]. Microbiological analyzes are carried out in the bacteriology laboratory of the Mohamed Boudiaf hospital, Isolated strains are identified through analysis based on standard microbiological techniques.

Tolerance of characterized strains to NaCl: We carried out this test to determine the optimum growth of the isolated strains and to see the influence of different NaCl concentrations according to their behavior. The isolated strains were tested for their tolerance to NaCl. Different concentrations (7.5%; 10%; 12.5%; 15%; 17.5%; 20% and 35%).

And we also carried out the test of antagonistic activity of bacterial strains with two referenced fungi, to study an important ecological, biotic factor which is the competition.

3. Results and Discussion

3.1. Results of physical and chemical analyses:

The samples of water taken from different sampling points show a T°C varied between 8°C and 10°C. The evolution of the temperature of a salt lake or chott remains linked to local conditions such as the regional climate, the topography, the duration of the sunshine, the flow and the depth [8].

The recorded pH of sample 3 is neutral varied between 6.6 to 7.2 while the pH of sample 1 and sample 2 tends towards alkalinity with values 7.55 and 8.39 successively.

The electrical conductivity (EC) of the water sample is between 2.4 and 6 ds/m, these results which allow us to qualify the water of the chott of Aïn El-Beida from salty to very salty (hyper salty) [9].

Identification involves a series of steps, most often following one another in a specific order. It is carried out based on standard microbiological techniques such as morphological and biochemical analysis (Gram staining, catalase and oxidase) and finally by the use of an API 20 E identification system. Concerning the bacterial density the values vary between 0.27×10^4 and 2.236×10^4 for the isolated strains. It is concluded that the microbial density of the chott water of Aïn El-Beida is abundant and diversified. For the results of the strains of the water samples of the Chott of Ain Elbaida are presented in Table 1.

Table 1. Results of the identification of strains isolated from the chott of Aïn El-Beida.

Isolats Numbers	Bactéria name
1	-
2	-
3	-
4	-

5	<i>Escherichia coli 2</i>
6	<i>Flavobacterium meningosepticum</i>
7	<i>Shigella spp</i>
8	-
9	-
10	-
11	-
12	<i>Escherichia coli 2</i>
13	<i>Aeromonas salmonicida</i>
14	-
15	-

- : Not identified

3.2. Tolerance of bacterial strains to NaCl

In the context of the characterization of isolated bacteria, we tested NaCl tolerance. The strains that have a high tolerance to NaCl are halophilic bacteria, which explains their presence in the chott of Aïn El-Beida. Salinity is considered an abiotic factor and may even be a limiting factor for the growth of these halophilic bacteria, as well as certain environmental conditions such as physico-chemical parameters such as pH, electrical conductivity and temperature influence the growth of microorganisms.

Table 2. The results of the strains at different NaCl concentrations.

NaCl concentration							
Isolat	7.5%	10%	12.5%	15%	17.5%	20%	35%
S1	+	-	-	-	-	-	-
S2	+	+	+	+	+	-	-
S3	+	+	+	+	+	+	+

S4	+	+	+	+	+	+	+
S5	+	+	+	+	+	+	+
S6	+	+	+	+	+	+	+
S7	+	+	+	+	-	-	-
S8	+	+	+	+	+	-	-
S9	+	+	+	+	+	-	-
S10	+	+	+	+	+	+	+
S11	+	+	+	+	+	+f	-
S12	+	+	+	+f	-	-	-
S13	+	+	+f	+f	-	-	-
S14	+	+	+	+	+	+	-
S15	+	+	+	+	+	+	+

+: Presence of growth; -: Absence of growth; +f: Low growth

All the strains studied tolerate a concentration of NaCl up to 15% except the strain (S1), six strains tolerate a concentration of 35% of NaCl. The tolerance of the strains is different for the 17.5% and 20% concentrations. The strains (S3, S4, S5, S6, S10, S11, S15) relatively tolerate high salt concentration of 20%-35%, and can be classified as euryhaline species, they are halophilic bacteria, as has been suggested by some authors [10]; [11]. By comparing our results with the work of including the identification of the LMB3981 strain in Lake El Goléa, where the NaCl concentration is equal to 25% (w/v). Regarding strains S2 to S15, the latter developed normally at NaCl concentrations of 5% to 17.5%, similar results are found in the hypersaline lake of Bakhtegan, located in southern Iran where they were able to identify the strain AF-2004 [12]. The tolerance of S2 to S15 strains shows that they can grow in environments where the NaCl concentration varies between 5 and 15%. The work of [13] Baati et al, 2009, at the Sfax-Tunisia salt station, revealed 40 halotolerant strains, the majority of which developed optimally between 5-15% salt, at 37°C and at pH 7.

3.3. Test for the antagonistic activity of bacterial strains

Among our objectives to highlight the ecological factors of our bacteria selected in the waters of the chott of Aïn El-Beida, to see their interactions with the biocenosis the environment, we chose

two fungi in order to evaluate the microbial activity of our strains which have been tested by the cross-streak method.

The fungus chosen for the antifungal test are referenced:

- *Aspergillus carbonarius* (Ac) which is a telluric fungus that produces toxic substances (mycotoxins).
- *Penicillium sp* (p).

The results obtained after an incubation period of 48 hours for the fungi, by measuring the zones of inhibition in millimeters between the border of the characterized strain and that of the target strain (Fungi-test).Show in figure 3.

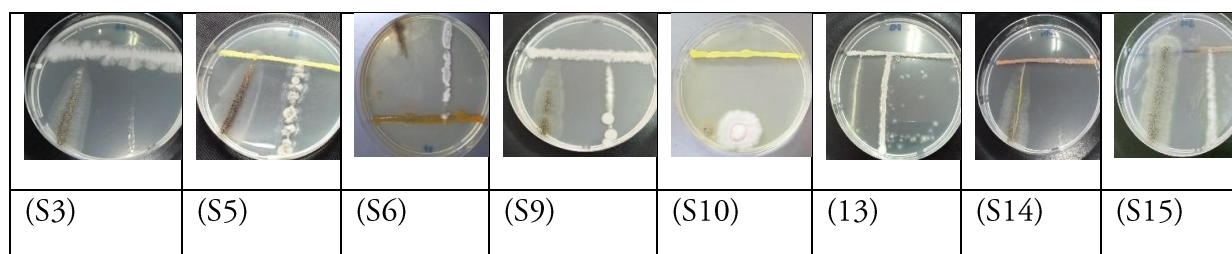


Figure 3. Antifungal activity of strains (S3,S5,S6,S9,S10,S13,S14,S15).

To the left Ac: *Aspergillus carbonarius*

To the right P: *Penicillium sp*

The S3 strain inhibits the growth of *Aspergillus carbonarius* (Ac) and *Penicillium sp* (P) as shown in figure 3.

The results of the antifungal activity of 15 isolated strains are presented in the table 3.

Table 3.Antifungal action of 15 selected isolates against 2 target fungi.

Muting distance Table 3.Antifungal action of 15 selected isolates against 2 target fungi. (mm)		
Isolat	<i>Aspergillus carbonarius</i> (Ac)	<i>Penicillium sp</i> (p)
S1	30	22
S2	35	35
S3	05	20

S4	20	27
S5	00	00
S6	40	00
S7	27	27
S8	50	40
S9	02	00
S10	30	20
S11	45	35
S12	40	35
S13	00	00
S14	00	-
S15	00	00

-: Absence

The results obtained clearly show that the antifungal action differs from one strain to another depending on the test fungi (show in the table 3).

For the antifungal activity of isolate S6 is considerable with the fungus *Aspergillus carbonarius* (Ac), on the other hand we note no antifungal activity against *Penicillium sp* (p).

The inhibition of fungal growth is mainly due to competition between the strains tested and the fungi used. *Aspergillus* produces a large number of spores which makes it more competitive, as has been suggested by some authors [14]. Competition between microorganisms also takes place for nutrients and according to [15] who showed that *Aspergillus* contains the element of competition for the acquisition of iron.

- For strain 3 and strain 9 showed resistance against *Aspergillus carbonarius* with an inhibition zone of 05mm and 02mm successively.

- S6 and S9 inhibited only *Aspergillus carbonarius* with an inhibition zone of 40 mm and 02 mm successively.

- The result of the antifungal activity showed the ability of S8 to strongly inhibit the growth of *Aspergillus carbonarius* (Ac) and *Penicillium sp* (p) with an inhibition zone of 50 mm and 40 mm successively.

Strains that show good resistance against fungi can be used in biological control against fungal contamination of dates caused by *Aspergillus carbonarius* (Ac) and *Penicillium* sp (p), the latter two were the most frequently isolated on dates [16](Abekhiti et al, 2013).

Conclusion

The isolated bacteria have a strong ecological valence and are euryhalines, and the results of the tests for NaCl tolerance and antagonism show that the bacteria chosen from the Chott of Ain Elbaida are tolerant, competitive, and resistant. The Chott of Ain El Baida is an ecosystem that is a part of the wetlands. Our ecological niche (the chott of An El-Beida) in the Ouargla region was able to provide us with some fascinating data according to this work on several bacteria. To better understand the biotic and abiotic factors that are directly and indirectly influencing the characterization of these bacteria, which are crucial for understanding the interactions between their biotope and their biocenosis, we conducted a variety of physico-chemical and microbiological analyses. We deduced from the physico-chemical analyses (T, pH, and EC) that the water in our samples had a temperature range of 8°C to 10°C. We discovered that our isolates, which can be categorized as euryhaline species, could tolerate relatively high salt concentrations during the various tests that were conducted on the strains that are selected and purified. It should be noted that some of our strains have a power of tolerance in NaCl up to 35%. The results of the antagonistic activity show that particular strains under study inhibit particular target fungi. The inhibitory capacity of strain 8 is highly effective against the intended fungus. S6 and S9, however, only inhibited *Aspergillus carbonarius*. To better understand the relationships between microorganisms and their natural environment, this work brings up a variety of viewpoints in these understudied ecosystems.

Conflicts of interest

The authors declare no conflicts of interest regarding the publication of this paper.

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