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Physicochemical and Microbiological Characterization of a Dry Soil in the Interface Steppe-Saharan Region in Southwest of Algeria

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ABSTRACT

The interface region steppe-Saharan, where the study was conducted is pastoral where the main activity is sheep production. The strong anthropological pressure has resulted in a reduction of the plant's potential, inducing a fragile ecosystem promoting more increased desertification. This work was undertaken in six stations distributed along a north-south transect. The physicochemical analyzes of these stations indicated a skeletal nature of the soil, sandy and limestone with CaCO₃ amount greater than 20, a pH ranging from 8.10-8.70 and a water retention capacity between 17.40-36%. The analyzes have also shown that the studied soils are rich in organic matter with a ratio greater than 4%, with a C/N ratio lower than 12. As for microbiological analyzes, they showed a bacterial presence varies that greatly from one station to another predominantly of *Pseudomonas* sp., *Staphylococcus* sp., *Clostridium sulphite*-reducing, *Enterobacteria* and *Streptococci* sp.

Key words: Ecosystem, microbiological analysis, physicochemical parameters, steppe-saharan

INTRODUCTION

The Algerian steppe has an area of 20 million ha. It is located on the edge of the northern Sahara (Brague-Bouragba *et al.*, 2007). It has a geographical entity differentiated by the harsh climate, the nature of its hydrology and particularly by the representative vegetation. The investigations undertaken *in situ* by Josa *et al.* (2011), allowed the identification of 137 plant species representing almost all plant families distributed in this area.

The phenomenon of the "regression ecosystem" (Peltzer *et al.*, 2010; Wardle *et al.*, 2004) concerns largely microbial communities and their effects on soil chemistry, nutrient availability and vegetation (Wardle *et al.*, 2004; Chadwick *et al.*, 1998).

In this study, the soil particle size was investigated. It involved coarse and fine sands, coarse and fine silts and clay.

This is because, aggregate stability is the main property of erosion control, runoff (Barthes *et al.*, 2000; Barthes and Roose, 2002) and plant growth (Gawlik *et al.*, 1999; Vdovic *et al.*, 2010) in semi-arid regions (Dunne *et al.*, 1991).

The main physicochemical porosity, moisture content, pH and electrical conductivity; involved in plant growth (Fan and Yang, 2007; Medina *et al.*, 2012) have also been studied.

In parallel, a biological soil analysis was conducted. This concerned the Enterobacteriaceae, *Staphylococcus*, *Clostridium sulphite*-reducing Streptococci, *Pseudomonas* and *Azotobacters* present in the soil. This fauna is important because they are involved in geochemical cycles and soil quality through the extracellular enzymes produced during the degradation of the organic matter complex (Sinsabaugh *et al.*, 1993).

As for the chemical analysis, major elements K, Na, P, total Ca, organic matter (Tagliavini *et al.*, 2005) and the C/N were analyzed. According to Pandey *et al.* (2011) and Wong *et al.* (2010), a high salt concentration inhibits the growth of microorganisms and therefore that of plants; causing land degradation (Nannipieri *et al.*, 2012) and a physicochemical change resulting in a significant loss of soil fauna (Singh *et al.*, 2012; Yuan *et al.*, 2007).

The study concerned the soil particle size, the main physicochemical parameters, the microbiological soil analysis, the major chemical elements and the organic matter. It was conducted in six stations located in the interface Saharan-steppe region, an arid bioclimatic zone of the El Bayadh in southwest Algeria. These stations were distributed along a north-south transect.

MATERIALS AND METHODS

Presentation of the study field and choice stations: The territory of the province of El Bayadh covers a total area of 6,642,039 ha, of which 5,704,445 ha includes steppe rangelands and pre-Saharan areas. The average rainfall is relatively low at 326 mm year⁻¹ (Josa *et al.*, 2011). The sampling sites were selected to cover the types of dominant and representative soils of the region. They are represented by six stations; ST1, ST2, ST3, ST4, ST5 and ST6 (Table 1).

Collection of soil samples: On each plot, six soil samples were carried out between 0 and 5 cm of depth, according to the Baize (2000) method. Next they were mixed to obtain a composite sample. The samples were stored in coolers at 4°C. They samples were used for the physicochemical and microbiological analyzes in the laboratory.

Physical and chemical analysis: The particle size was determined by the method described by Aubert (1978). The porosity, moisture content, residual moisture, pH, electric conductivity, organic matter and carbon were analyzed by Petard (1993) method. The amount of assimilable phosphorus was realized by Olsen Method, the nitrogen by the Kjeldahl method, the total calcareous content by Bernard's calcimeter.

Analysis and enumeration of prokaryotes: Ten grams of soil samples were diluted in 90 mL of physiologic and sterile water, then stirred with a vortex. From this stock solution, a

series of dilutions were carried out appropriately. One hundred microliter of each dilution was used for the different analyzes provided in this work.

A non-selective nutrient agar medium was used for the search of the total flora. By contrast, selective media glucose agar with bile compound, Baid parker, Meat-Liver, Hagar bile esculin of Cetrinide and synthetic medium were used, respectively for the detection and enumeration of Enterobacteriaceae, staphylococci, of sulphite-reducing *Clostridium*, *Streptococcus*, *Pseudomonas* and *Azotobacters*.

Incubation of microorganisms was carried out in Petri dishes at 37°C for 24-72 h. Macroscopic and microscopic examinations and conventional biochemical tests for bacterial identification were performed according to the Method in Bergey's Manual (1986).

Analysis and enumeration of eukaryotic: Eukaryotic existing soil fungi were studied. Incubation of the samples containing the fungi was performed at 25°C for 3-5 days in Petri dishes containing the medium supplemented with citric acid, sulfated streptomycin and chlortetracycline hydrochloride with 250, 100 and 50 mg L⁻¹, respectively. Enumeration of viable bacteria was expressed in colony forming units (CFU) per gram of sample using the following formula (Beraud, 2001):

$$\text{Log CFU g}^{-1} \text{ of sample} = \frac{\text{Number of colonies}}{\text{Dilution} \times \text{Volume seeded}}$$

Statistical analysis: Statistical analyses were performed using SPSS Version 17 software. All experimentations were performed in duplicate. The results are expressed as Mean±standard deviation ESM. The Pearson r coefficient was used to study the correlation between organic biomass and other factors. Because, it measures adequately the linear relationship between these two variables.

The value of P or sig. (bilateral) introduced by Gibbons and Pratt (1975) and used by several authors (Bezeau and Graves, 2001; Cashen and Geiger, 2004; Maddock and Rossi, 2001; Paul and Plucker, 2004) to confirm the actual links between the factors studied. Therefore, it implies a causal relationship between the two variables.

RESULTS AND DISCUSSION

Grain size: Table 2 showed a sandy texture of ST1, ST2, ST3, ST5 and ST6. However, the St4 showed a sandy-loam texture.

It should be noted that sandy soils are often dry, nutrient-poor and very draining. They have an unstable structure. They are, therefore, susceptible to wind erosion. As for sandy loam (ST4), the soil is relatively more fertile and retains moisture.

Total porosity: According to Chong (2005) and Vaughn *et al.* (2011), the total porosity of the soil is between 50 and 85%. In

Table 1: Locations of study sites

Stations	Geographic	Coordinates (m)
Haoudh (ST1)	1° 09' 02,2'' E; 33° 55' 30,4'' N	1294
Laguermi (ST2)	1° 12' 24,0'' E; 33° 34' 52,1'' N	1303
Ghassoul (ST3)	1° 09' 02,5'' E; 33° 31' 52,6'' N	1376
Stitena (ST4)	1° 13' 61,1'' E; 33° 41' 68,6'' N	1457
Hadjrat derissa (ST5)	1° 13' 61,1'' E; 33° 13' 61,0'' N	1400
Krekda (ST6)	1° 09' 02,2'' E; 33° 31' 52,9'' N	1366

Table 2: Results of physico-chemical analyses of soil in study stations

Variables	ST 1	ST 2	ST 3	ST 4	ST 5	ST 6
Grain size						
CS	45.35	40.98	10.94	33.75	33.75	41.64
S	44.38	49.43	80.04	49.39	49.39	46.58
CS	2.50	3.52	3.52	13.16	5.31	5.00
FS	1.50	3.50	1.50	2.03	5.20	2.00
C	1.00	2.00	1.00	1.00	0.80	3.00
Porosity	41.50	41.50	46.03	46.79	57.35	52.83
Water retention (%)	17.47	23.24	23.06	26.04	36.02	32.38
Moisture (%)	0.90	1.00	2.40	2.50	2.50	2.60
pH (H ₂ O)	8.60	8.70	8.20	8.70	8.10	8.20
EC (mmhos/cm)	1.152	1.222	1.502	1.035	1.2212	1.515
Organic matter (%)	5.00	24.15	4.65	35.85	28.25	8.45
C/N	11.60	11.70	11.74	11.64	11.64	11.69
Limestone (%)	35.96	37.00	30.01	20.59	26.46	25.90
K (%)	4.00	4.00	4.40	4.90	11.89	4.40
Na (%)	0.05	0.06	0.05	0.07	0.125	0.05
P (ppm)	5.00	8.00	6.00	3.00	8.00	10.00

CS: Coarse sand, S: Sandy, CS: Coarse silt, FS: Fine silt, C: Clay, EC: Electrical conductivity

Table 3: Test pearson correlations (r) and the significance test of the slope or Sig. (bilateral) (P) between the different factors with number of stations N = 6

Variables	RH	OM	C/N	Na	P	BM
RH correlation	0.206	0.279	0.334	0.106	-0.251	
pearson (r)						
Sig. (bilaterale) (P)	1	0.695	0.593	0.518	0.842	0.632
N	6	6	6	6	6	6
OM correlation	0.206		-0.258	0.635	-0.267	0.836
pearson (r)						
Sig. (bilaterale) (P)	0.695	1	0.622	0.176	0.609	0.38
N	6	6	6	6	6	6
C/N correlation	0.279	-0.258		-0.322	0.388	-0.322
pearson (r)						
Sig. (bilaterale) (P)	0.593	0.622	1	0.534	0.447	0.534
N	6	6	6	6	6	6
Na correlation	0.334	0.635	-0.322		0.097	0.213
pearson (r)						
Sig. (bilaterale) (P)	0.518	0.176	0.534	1	0.854	0.686
N	6	6	6	6	6	6
P correlation	0.106	-0.267	0.388	0.097		-0.322
pearson (r)						
Sig. (bilaterale) (P)	0.842	0.609	0.447	0.854	1	0.534
N	6	6	6	6	6	6
BM correlation	-0.251	0.836	-0.322	0.213	-0.322	
pearson (r)						
Sig. (bilaterale) (P)	0.632	0.38	0.534	0.686	0.534	1
N	6	6	6	6	6	6

RH: Residual moisture, OM: Organic matter, BM: Biomass, C/N ratio, Carbon products: Nitrogen products

our case (Table 2), ST5 and ST6 have a relatively high porosity. Therefore, they have a water retention capacity that is higher compared to other stations.

Water retention and moisture: The results (Table 3) show that the moisture content varies from one station to another. However, it is higher in stations ST3, ST4, ST5, ST6. However, in ST1 and ST2, the rate is about 1%. These findings may be related to the vegetation cover. Because, the roots play an important role in the exchange and maintenance of plant-animal symbiosis in the soil (Choi *et al.*, 2010).

pH: The pH of the soils studied is between 8.2 and 8.7 (Table 2) within 6 stations. This high pH is according to Mengel and Kirkby (2001) caused by an absence of ammoniacal nitrogen. A qualitatively high pH affects the solubility of some elements; thus influencing their absorption Raven *et al.* (2010). On the other hand, in more acidic soils, nitrification is greater (Cookson *et al.*, 2004; Tong and Xu, 2012; Norton and Stark, 2011; Serjeant and Dempsey, 1979). According to Babiker *et al.* (2004), De Paz and Ramos (2004), Albregts and Howard (1980), Urrestarazu *et al.* (2008) and Abad *et al.* (2001), a pH between 5.5 and 6.5 allows full absorption of microelements.

Electrical conductivity: The electrical conductivity of soils in the study area varies between 1.15 and 1.5 dS m⁻¹ (Table 2). We deduce, while moderately saline soils are studied. If the level of salinity is less than 2.0 dS m⁻¹, the effects on the plant's growth are negligible (Saied *et al.*, 2005; D'Anna *et al.*, 2003). Caruso *et al.* (2011) and Baize (2002). The ideal values are between 2.18-2.34 dS m⁻¹ (De Pascale *et al.*, 2001; Keutgen and Pawelzik, 2007; Choi and Latigui, 2008; Skiredj, 2005). Greater than those above mentioned values can cause high salinity consequently, causing phytotoxicity (Latigui, 1992; Latigui *et al.*, 2011).

Organic matter and C/N: The studied soils are rich in organic matter (Table 2). This reflects the abundance of vegetation cover. Generally, the organic status is evaluated by measuring the total organic carbon concentration (TOC) and Total Nitrogen (TN) in the soil. This is due to the estimate of the N/C ratio that shows values lower than 20; optimal for the mineralization of organic matter values (Genot *et al.*, 2011). The higher levels of organic matter associated can be also associated with an improvement of the structure, facilitation of infiltration, an increase in water retention capacity and the ability to resist erosion (Raven *et al.*, 2010).

Limestone: The contents of total calcium in the soils studied are between 20.60-37% (Table 2). Thus, it appears that the soil in the study areas are calcareous like most Algerian soils. It should be noted that according to Benchetrit (1959) 70-80% of the soil of Algeria is slightly to moderately filled with limestone horizons deep in the soil versus superficially.

Potassium and sodium: For the rate of K and Na, the highest values were found in ST5 with 11.89 and 0.125% (Table 2), respectively. This value appears to be due to the abundance and/or the type of vegetation cover (Genot *et al.*, 2011). A high concentration of Na inhibits the nitrogen cycle, thus influencing the microbial biomass, enzyme activity and nitrification (Tong and Xu, 2012; Matsushima *et al.*, 2009).

Phosphorus: In this study, ST6 contains relatively more of the available P (Table 2). This value appears to be due to the

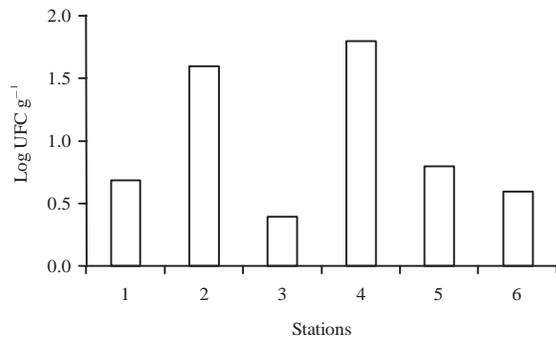


Fig. 1: Total microbial biomass for each station

density of vegetation cover or to the herbaceous dominant type in the region (Balesdent, 1996). An inappropriate concentration of P causes a profound change in the aerial (De Groot *et al.*, 2001) and root parts of the plants. A low concentration of P is mainly due to mineral weathering and leaching (Izquierdo *et al.*, 2013).

2 laboratory tests

Microbiological analysis: Microbiological analysis (Fig. 1) showed the presence of a complex and diverse microbial biomass. It is relatively denser in the soils of ST4. This relative growth of microbial flora is related to the low electrical conductivity negligible in this threshold (Table 1), the richness of soil organic matter with water holding capacity and the highest low limestone. These factors promote microbial growth (De Pascale *et al.*, 2001; Raven *et al.*, 2010; Kuroiwa *et al.*, 2011).

Parameters such as pH and nutrient availability are known to be important factors for the structure, diversity and function of communities both plant and microbial (Grayston *et al.*, 2004; Lauber *et al.*, 2009; Marschner *et al.*, 2004).

Bacterial biomass is a sensitive indicator of a long-term decline of soil organic matter resulting from a disturbance of a natural ecosystem. The organic matter regulates the biological activities and contributes to the diversity and complexity of the soil. It is, moreover, a large reserve of nutrients that will be available to the plant (Genot *et al.*, 2011).

Fungal analysis: A comparative analysis showed that the rate of fungi growth was lower than those of bacteria (Fig. 2).

According to Roux (2005), the increase in humidity stimulates bacteria. A slightly alkaline pH is optimum for growth of bacteria whereas the fungi prefer a low pH from 3-5.

Distribution of bacterial flora: The microscopic and biochemical identification of microorganisms isolated showed the presence of Enterobacteriaceae, *Staphylococcus* sp., *Streptococcus* sp., *Pseudomonas* sp. These represent a large fraction of the bacterial community particularly in ST5. They are found in all walks of life, especially on the root systems of

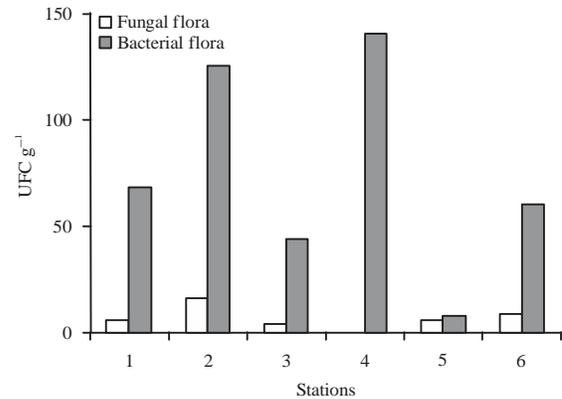


Fig. 2: Distribution of bacterial and fungal flora in each station

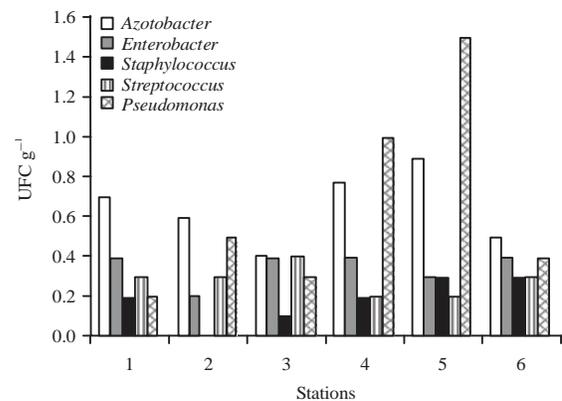


Fig. 3: Bacterial breakdown of the six stations

plants (Haas and Keel, 2003). Their predominance in soils can be explained by a high density of vegetation (Josa *et al.*, 2011; Chapin III, 1980), particularly in ST5 (Fig. 3), where the canopy is 75-80% (Josa *et al.*, 2011).

Moreover, porosity, aeration and depth have, firstly, the growth and development of the genus *Pseudomonas* sp., strict and also inhibit, also the proliferation of aerobic bacteria *Clostridium sulphite* (Roux, 2005).

Since the study area is an integral part of transhumance routes, the presence of staphylococci and streptococci, ubiquitous bacteria may have an animal origin. The abundance of nitrogen-fixing bacteria *Azotobacter* sp. is found in the soil of ST5, particularly in the rhizosphere of *Pinus halpensis*.

Distribution of the fungal flora: The analysis shows the predominance of fungal flora in ST2 while, in ST4 it is devoid. This explains the decrease in phosphorus in the later (Fig. 2). According to Bolan (1991), fungi improve the collection and transport to the plant very little mobility mainly phosphorus nutrients. They increase tolerance to drought and reduce the effects of pathogenic infections. In addition, positive interactions were found between mycorrhizal interactions.

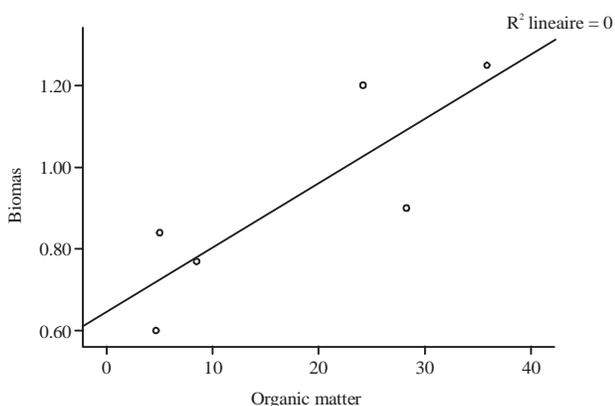


Fig. 4: Presentation of regression line

Fungi and soil bacterial communities: Interpretation of the results (Table 3) was made by combining the Pearson correlation (r) and tests for significance of the slope (p).

Pearson correlation (r) allows us to determine if the relationship between the 2 factor is perfect (1), very high (0.85), high (0.5-0.8), medium (0.2-0.5), low (0-0.2) or zero (0). However, Sig. (bilateral) (p) allows us to determine if this relationship is significant and not due to chance. Recall that if the Sig. or p -value is greater than 0.5 we conclude that the correlation expressed by Pearson's R is due to chance. However, if P is less than 0.5, the relationship between the two factors is significant.

In our case, we deduce that the Mo-Na correlation ($r = 0.635$, $p = 0.176$) is strong and that there is indeed a relationship between the two factors.

The correlation C/NP ($r = 0.388$, $p = 0.447$) is moderate, but really exists.

The correlation BO-MO ($r = 0.836$, $p = 0.38$) is very strong and really exists (Table 3 and Fig. 4).

CONCLUSION

Parameters such as pH and nutrient availability are known to be important factors in the structure, diversity and function of communities of both plant and flora.

This approach seems to be an interesting contribution to the study of semi-arid soil with a much diversified native flora, with the exception in ST3 whose land has the introduced *Atriplexe canescens*.

In light of our results, we can infer that the soils of our study sites are sandy and skeletal. By contrasts, the soil of ST4 is sandy loam, limestone and rich in organic matter with an alkaline pH favoring the development of bacterial flora compared to the fungal flora.

Through correlation tests, this diversity is not only related to the physicochemical parameters of the soil but also to the existing microbial community and its frequency. Microbiological characterization revealed a diversity in bacterial flora and fungal flora. The bacterial flora includes

Enterobacteriaceae, *Staphylococcus* spp, *Streptococcus* sp, the sulphite-reducing *Clostridium*, with a predominance of *Pseudomonas* sp.

Steppe soils are fragile and are easily eroded. Nevertheless, there is a balance in the ecosystem, especially between the physicochemical and microbiological parameters supporting a significant vegetation cover or more or less discontinuous, that is essential for maintaining pastoral activity characteristic of this region. This will allow periodic monitoring of these indicators. Because, they inform us about the soil's ability to withstand the anthropological pressure. They will also, help ecologists choose species for highly sensitive areas that can adapt to these conditions in rehabilitation programs.

The perspective is to set up a database in a Geographical Information System (GIS). It would integrates multiple data sources physical, chemical, microbial, climate, water and socio-economic in order to monitor and intervene in real time to minimize the phenomena of steppe desertification.

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