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Chapter 6

Biodiversity of Amphipoda Talitridae in Tunisian Wetlands

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Abstract

Although wetlands were remarkable habitats with their fauna and flora diversity, few studies have been devoted to the study of amphipod biodiversity in this ecosystem type. The amphipod communities of six wetland types belonging to 117 stations were studied with respect to species composition, abundance and their relationship with environmental parameters. Amphipods were collected during spring. At each station, eight quadrats of 50 × 50 cm^2 were randomly placed. Animals were preserved in alcohol at 70°C. In the laboratory, the specimens collected were identified and counted. Physicochemical parameters (organic matter, particle size, heavy metals) of sampled soils were determined. The results showed that the highest species richness was observed in lagoons with the presence of eight species namely *Orchestia montagui, Orchestia gammarellus, Orchestia mediterranea, Orchestia stephenseni, Orchestia cavimana, Platorchestia platensis, Deshayesorchestia deshayesii* and *Talitrus saltator*, whereas in the hill lakes and dams banks, no specimens were collected. The biodiversity of amphipod species depends on climatic (temperature, humidity) and edaphic (organic matter, particle size, heavy metals) factors.

Keywords: Tunisia, wetlands, neuro-inflammation, Amphipoda, diversity, environmental factors

1. Introduction

In the Mediterranean, there was a high diversity of wetlands (lagoon, lake, sebkha, wadi, hill reservoir and dam) that were of great importance in conservation of biology. They were considered among the most biologically diverse and productive ecosystems [1]. They offer a wide variety of natural habitats for plants and aquatic animals as well as semi-terrestrial and terrestrial species. The interactions of biological (plants, animals, microorganisms, etc.) and physicochemical components (granulometry, temperature, humidity, etc.) of wetlands enable them to perform many ecological functions such as shoreline stabilization and water purification. Lacaze [2] mentioned that lagoon wetlands harbour a diverse fauna, but were threatened by intense anthropogenic exploitation and pollution. As they receive continental freshwater from their catchment area, many lagoons have been subjected to severe degradation of water quality caused by pollution and/or eutrophication [3]. In Tunisia, semi-closed shallow lagoons were among the most sensitive areas to environmental stresses [4, 5].

Among wetlands, sandy beaches were more studied and characterized by the presence of a large number of invertebrates. Talitridae amphipods were among the most dominant invertebrates living on wetlands [6]. These talitrids play an important role as decomposers of organic matter and were considered as potential bio-indicators of sandy beaches quality [7, 8]. This role was estimated using genetic approach, behavioural approach as well as reproduction and spatio-temporal distribution studies [9–32].

In Tunisia, amphipod communities inhabiting wetlands bank, other than sandy beaches [21–23, 31–35] have not received much attention. Through this study, we propose a description as exhaustive as possible of the biodiversity of these communities taking into account geographical, climatic and edaphic specificities. More specifically, we addressed the following questions: (1) Does the diversity of Talitridae amphipods follow a north-south cline? (2) Is the correlation between specific diversity and wetlands type is significant?

2. Materials and methods

2.1. Study site

This study focuses on wetlands that consist of permanent or temporary areas of fresh or brackish water and adjacent lands. They include all wadis, chotts, lagoons, hill lakes, sebkhas and dams. The majority of these areas, several of which were of international importance, were found in the north, particularly near the coast. In this study, 117 stations namely *lagoons* (a stretch of salt water partially or completely separated from the open ocean by barriers of sand or coral distributed along the Tunisian coasts), *lakes* (a body of relatively still freshwater of considerable size, localized in a basin that was surrounded by land and most of them were fed and drained by rivers and streams), *sebkhas* (North African vernacular name for a shallow, salty depression. It was a common wetland type especially in semi-arid and arid climate), *wadis* (a natural stream of water of fairly large size flowing in a definite course or channel or series of diverging and converging channels), *hill lakes* (distinguished by a height >10 m and a volume >1 million m^3) and *dams* (characterized by a reservoir volume more than 3 million m^3 and a height of 15 m) were prospected (**Table 1**).

2.2. Sampling methods and laboratory procedures

Quantitative samples of amphipods were taken in spring of 2008, 2009 and 2010 in the early morning hours using quadrates method [36, 37]. In the bank of each site, eight quadrates of 50×50 cm^2 were randomly placed. The content of each quadrat (7 cm depth) was placed in an

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Stations	Wetland type	Governorate	GPS	Sediment type
1. Bizerte	Lagoon	Bizerte	37°13′8″N/009°55′1″E	Loamy sand
2. El Bcherliya	Lagoon	Bizerte	37°10′03″N/010°09′57″E	Loamy sand
3. Ghar El Melh Old harbour	Lagoon	Bizerte	37°10′04″N/010°11′40″E	Loamy sand
4. Boughaz	Lagoon	Bizerte	37°10′09″N/010°13′12″E	Sandy loam
5. Sidi Ali Mekki	Lagoon	Bizerte	37°09′50″N/010°14′45″E	Fine silt
6. Tunis North lagoon	Lagoon	Tunis	36°48′01″N/010°12′27″E	Loamy sand
7. Tunis South lagoon	Lagoon	Tunis	36°47′59″N/010°12′26″E	Sandy
8. Korba lagoon	Lagoon	Nabeul	36°38′12″N/010°54′11″E	Loamy sand
9. Tazarka lagoon	Lagoon	Nabeul	36°32′20″N/010°50′38″E	Sandy-clay-silt
10. Bhiret El Biben	Lagoon	Medenine	33°15′57″N/011°08′28″E	Sandy
11. Ichkeul	Lake	Bizerte	37°06′37″N/009°41′21″E	Loamy sand
12. Bouhnach	Lake	Ariana	36°58′57″N/010°08′56″E	Sandy loam
13. Majin Chitane	Lake	Bizerte	37°09′07″N/009°05′54″E	Sandy-clay-loam
14. El Ouafi	Sebkha	Bizerte	37°09′22″N/010°13′38″E	Sandy Silt
15. Raoud	Sebkha	Ariana	36°55′57″N/010°10′48″E	Clay
16. Ariana	Sebkha	Ariana	36°56′53″N/010°11′4″E	Sandy clay
17. Kalaat Andalous	Sebkha	Ariana	37°05′06″N/010°10′16″E	Clay
18. Sliman	Sebkha	Nabeul	36°42′02″N/010°27′37″E	Sandy
19. Maâmoura	Sebkha	Nabeul	36°28′2″N/010°48′21″E	Sandy
20. Sidi Khlifa	Sebkha	Sousse	36°14′20″N/10°26′15″E	Clay
21. Assa Jriba	Sebkha	Sousse	36°0′46″N/10°25′36″E	Clay
22. Halk El Menzel	Sebkha	Sousse	36°0′23″N/10°27′15″E	Loamy sand
23. Sousse	Sebkha	Sousse	35°47′45″N/10°38′48″E	Sandy silt loam
24. Monastir	Sebkha	Monastir	35°46′21″N/10°46′47″E	Loamy sand
25. Argoub	Sebkha	Gabès	33°38′20″N/10°16′56″E	Clay
26. Khalfallah	Sebkha	Medenine	33°26′59″N/010°56′32″E	Clay
27. Gorgabiya	Sebkha	Medenine	33°23′45″N/10°54′54″E	Sandy
28. Moknine	Sebkha	Monastir	35°37′13″N/10°55′17″E	Clay loam
29. Gargour	Sebkha	Sfax	34°37′32″N/10°38′22″E	Sandy clay
30. Sidi El Hani	Sebkha	Sousse	35°32′14″N/010°18′35″E	Sandy Silt
31. Kalbiya	Sebkha	Kairouan	35°54′25″N/010°17′08″E	Silty
32. Metbasta	Sebkha	Kairouan	35°45′14″N/010°06′57″E	Silty
33. M′Habbil	Sebkha	Medenine	33°24′47″N/010°51′59″E	Clay
34. Kairouan	Sebkha	Kairouan	35°44′6″N/010°6′52″E	Silty
35. Mchiguig	Sebkha	Sfax	34°58′58″N/010°03′06″E	Sandy Silt

Stations	Wetland type	Governorate	GPS	Sediment type
36. Thrayaa	Sebkha	Gabès	34°10′10″N/010°00′47″E	Sandy silt loam
37. Gataaya	Sebkha	Kébili	33°41′44″N/008°53′44″E	Sandy clay
38. Jemna	Sebkha	Kébili	33°34′48″N/009°00′15″E	Clay
39. Blidette Sguira	Sebkha	Kébili	33°35′18″N/008°51′06″E	Sandy silt loam
40. Blidette Kbira	Sebkha	Kébili	33°34′27″N/008°51′37″E	Sandy silt loam
41. Guidma	Sebkha	Kébili	33°25′44″N/008°47′45″E	Sandy clay
42. Golaa	Sebkha	Kébili	33°31′18″N/008°57′26″E	Sandy clay
43. Zarzara	Sebkha	Kébili	33°31′07″N/008°56′30″E	Clay
44. El Korsi	Wadi	Bizerte	37°11′12″N/009°46′52″E	Sandy loam
45. Tinja	Wadi	Bizerte	37°10′10″N/009°45′26″E	Loamy sand
46. Lebna wadi Estuary	Wadi	Nabeul	36°38′58″N/010°54′57″E	Sandy loam
47. Khniss	Wadi	Monastir	35°43′13″N/010°48′57″E	Sandy
48. Lakaarit	Wadi	Gabès	34°06′29″N/009°58′55″E	Sandy
49. El Fared	Wadi	Gabès	33°44′59″N/010°12′31″E	Sandy-clay-silt
50. Majerda	Wadi	Bizerte	37°05′03″N/010°08′17″E	Loamy sand
51. Joumin	Wadi	Bizerte	37°0′37″N/009°41′59″E	Sandy
52. Sidi Bou Ali	Wadi	Sousse	35°58′8″N/010°27′20″E	Sandy
53. Hamdoun	Wadi	Monastir	35°46′51″N/010°40′48″E	Sandy loam
54. Zerkine	Wadi	Gabès	33°41′22″N/010°15′12″E	Sandy
55. Zigzaw	Wadi	Gabès	33°35′40″N/010°18′42″E	Clay
56. Zas	Wadi	Medenine	33°30′53″N/010°20′28″E	Clay
57. Koutine	Wadi	Medenine	33°26′34″N/010°23′9″E	Silty
58. Hessi Amor	Wadi	Medenine	33°21′47″N/010°37′14″E	Clay
59. Bouhamed	Wadi	Sidi Bouzid	33°18′6″N/010°44′5″E	Silty
60. Demna	Wadi	Gabès	33°56′27″N/010°1′35″E	Loamy sand
61. Maleh	Wadi	Gabès	34°0′2″N/009°59′57″E	Loamy sand
62. Widran	Wadi	Sfax	34°31′7″N/010°4′17″E	Clay
63. Zit	Wadi	Zaghouan	36°27′01″N/010°16′43″E	Sandy
64. El Harat	Wadi	Zaghouan	36°21′50″N/010°18′34″E	Sandy
65. Lassoued	Wadi	Siliana	36°24′20″N/010°12′37″E	Sandy loam
66. Sidi Hmid	Wadi	Zaghouan	36°24′21″N/009°58′56″E	Silty
67. Bouthiben	Wadi	Zaghouan	36°22′16″N/009°54′0″E	Sandy
68. El Kbir wadi	Wadi	Siliana	36°13′26″N/009°44′49″E	Loamy sand
69. Siliana	Wadi	Siliana	36°12′03″N/009°42′57″E	Sandy-clay-silt

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Stations	Wetland type	Governorate	GPS	Sediment type
70. El Kbir	Wadi	Siliana	36°07′11″/009°35′28″E	Sandy-clay-silt
71. Bargou	Wadi	Siliana	36°05′25″N/009°33′48″E	Loamy sand
72. Massouj	Wadi	Siliana	36°04′57″N/009°22′30″E	Fine silt
73. Saboun	Wadi	Siliana	35°52′11″N/009°11′37″E	Silty
74. Zguifa	Wadi	Siliana	35°45′55″N/009°01′22″E	Loamy sand
75. Raguey	Wadi	Jendouba	36°27′51″N/008°23′27″E	Sandy loam
76. Mazbla	Wadi	Jendouba	36°29′11″N/008°18′28″E	Sandy loam
77. El Maleh	Wadi	Ariana	36°58′41″N/010°09′55″E	Sandy loam
78. Lanj	Wadi	Jendouba	36°34′46″N/008°30′25″E	Sandy
79. Lahmam	Wadi	Jendouba	36°32′55″N/008°26′53″E	Loamy sand
80. Soufi	Wadi	Jendouba	36°29′20″N/008°23′49″E	Sandy loam
81. Menzel Tmim	Wadi	Nabeul	36°42′26″N/010°43′27″E	Sandy loam
82. El Widyen	Wadi	Nabeul	36°47′03″N/010°53′39″E	Sandy
83. Sliman	Wadi	Nabeul	36°41′36″N/010°28′53″E	Loamy sand
84. Lebna	Wadi	Nabeul	36°39′13″N/010°54′31″E	Sandy loam
85. Houith	Hill lake	Bizerte	37°4′59″N/009°58′5″E	Loamy sand
86. Morra	Hill lake	Bizerte	37°05′53″N/009°59′08″E	Sandy
87. Bnt Liba	Hill lake	Bizerte	37°05′52″N/009°59′08″E	Sandy-clay-silt
88. Ghar Ettine	Hill lake	Bizerte	37°04′02″N/009°15′53″E	Sandy
89. Sidi Daoued	Hill lake	Bizerte	37°03′14″N/009°23′47″E	Sandy
90. Khelifa wadi	Hill lake	Zaghouan	36°13′40″N/009°47′13″E	Sandy
91. Jetta	Hill lake	Siliana	35°59′44″N/009°26′48″E	Sandy
92. Ain Ben Ali	Hill lake	Siliana	36°03′47″N/009°17′35″E	Sandy
93. Zrab wadi	Hill lake	Siliana	36°02′8″N/009°16′54″E	Sandy
94. Khalsi	Hill lake	Siliana	35°57′10″N/009°10′32″E	Sandy
95. Jdaïda wadi	Hill lake	Siliana	35°53′53″N/009°11′12″E	Loamy sand
96. Ettal wadi	Hill lake	Siliana	35°53′20″N/009°10′54″E	Sandy loam
97. Ksayir Hamdoun	Hill lake	Siliana	35°48′10″N/009°03′57″E	Sandy
98. Ouled Ali	Hill lake	Siliana	35°50′58″N/009°09′31″E	Sandy
99. Zraybiya	Hill lake	Jendouba	36°28′25″N/008°21′29″E	Loamy sand
100. At 5km d'El Kssour	Hill lake	Kef	35°52′05″N/008°55′52″E	Clay
101. Bni Mtir	Dam	Jendouba	36°44′47″N/008°44′19″E	Sandy loam
102. Sidi Barrak	Dam	Béja	37°00′52″N/009°06′12″E	Sandy
103. El Hma	Dam	Ben Arous	36°35′16″N/010°18′24″E	Sandy clay
104. Bakbaka	Dam	Ben Arous	36°34′35″N/010°20′17″E	Loamy sand

Stations	Wetland type	Governorate	GPS	Sediment type
105. Bnt Jedidi	Dam	Nabeul	36°25′09″N/010°27′26″E	Sandy-clay-silt
106. Ermal wadi	Dam	Sousse	36°19′50″N/010°21′29″E	Sandy loam
107. Jneyhiya	Dam	Siliana	36°12′25″N/009°44′20″E	Sandy loam
108. Siliana	Dam	Siliana	36°07′57″N/009°21′14″E	Loamy sand
109. Lakhmas	Dam	Siliana	35°59′55″N/009°28′15″E	Sandy-clay-silt
110. El Gattar	Dam	Siliana	36°01′47″N/009°15′56″E	Sandy
111. Cheikh El Maïz	Dam	Siliana	36°01′15″N/009°15′8″E	Sandy
112. El Kharroub wadi	Dam	Siliana	36°01′43″N/009°15′8″E	Sandy
113. Mchaker wadi	Dam	Siliana	35°58′57″N/009°10′20″E	Sandy loam
114. Ermal	Dam	Siliana	35°49′21″N/009°07′33″E	Loamy sand
115. Mallègue	Dam	Kef	36°18′48″N/008°42′21″E	Sandy loam
116. Kasseb	Dam	Béja	36°45′36″N/009°0′5″E	Sandy
117. Ermal	Dam	Siliana	36°23′54″N/010°04′52″E	Loamy sand

Table 1. Localization of the studied stations.

individual bag, and then the animals were sorted by hand. Twenty minutes were devoted to each quadrat. Humidity and temperature of air and soil were measured *in situ* at each site. At the laboratory, amphipod specimens were preserved in 70% ethanol. Then, they were identified, counted and sexed. The identification of these species was carried out under Leica MS 5 binocular microscope, using the key of Ruffo [38].

2.3. Soil analysis

The particle size, organic matter and heavy metals of soil samples taken from 117 stations were analysed. Grain size distribution of these composite samples was analysed using different sieves in descending order (from 2 to 25 μm).

A subsample was brought to the inductively coupled plasma-mass spectrometry (ICP-MS) laboratory at University of Kiel and sieved to obtain the <250-μm grain size fraction which was then dried and milled [39]. Heavy metals were extracted from a 250-mg sample of powder with 10 mL 7 N nitric acid on a hot plate at 80°C (2.5 h). The solution was made up to 20 mL, centrifuged at 3500 rpm for 15 min, and the supernatant transferred to a 20-mL sample vial. The metals vanadium (V), chromium (Cr), manganese (Mn), cobalt (Co), nickel (Ni), copper (Cu), zinc (Zn), arsenic (As), cadmium (Cd), tin (Sn), thallium (Tl), lead (Pb), lithium (Li), rubidium (Rb) and strontium (Sr) were analysed by inductively coupled plasma-mass spectrometry (ICP-MS). Average analytical reproducibility was estimated from replicate analyses of some samples and was found to be better than 2% Relative Standard Deviation (RSD) (1 sigma relative standard deviation) for all elements. The accuracy of analytical results was monitored by analysing certified reference materials (CRM): GSMS-2 (marine sediment; Chinese Academy of Geological Sciences, PR China) and Reference material, coastal sediment (PACS-1) (coastal sediment;

National Research Council Canada (NRCC) Canada) as unknowns along with the samples. Organic matter content was determined by weighing before and after ashing at 450°C for 3 h at the University of Salzburg.

2.4. Data analysis

To compare the amphipod community structure among stations, different faunistic parameters were calculated using quantitative data such as species richness, relative species abundance, etc. Mean density of the amphipod community at each station and the mean density of each species at each station were expressed as number of individuals per m^2. Species diversity and evenness were calculated by the Shannon-Weaver index and Pielou's evenness index [40], respectively. The degree of similarity between sampling stations was evaluated using similarity cluster dendrograms. The analysis above was performed with the PRIMER software package [41]. Principal component analysis of amphipod distribution and site characteristics was performed using Xlstat software.

3. Results

3.1. Temperature, humidity, organic matter and grain size

Temperature (°C) and humidity (%) were measured *in situ* in different wetland types. The mean values for these two parameters varied between 22.453 ± 2.797°C in dams, 27.387 ± 5.289°C in sebkhas, 51.243 ± 18.627% in sebkhas and 65.50 ± 12.388% in lagoons (**Figure 1A** and **B**).

The percentage of organic matter differs between and within wetland types (**Figure 1C**). The highest values were observed in the banks of Bizerte lagoon (9.46%), Majin Chitane (12.23%), Halk Menzel (16.13%), Bargou wadi (20.66%), Ouled Ali (17.62%) and Kasseb (12.64%) (**Figure 1C**).

An heterogeneity in grain size nature was observed between stations ranging from sandy substrates, loamy sand, sandy loam, sandy silt, sandy-clay, silty clay, clay-loam, sandy-clay-silt, sandy-clay-loam, sandy-silt-loam, fine silt to clay substrates (**Table 1**).

3.2. Heavy metals

In the lagoon, the highest concentrations for the majority of heavy metals, vanadium, nickel, zinc, arsenic, cadmium, thallium and lead were recorded in the northern lagoon of Tunis. The bank of Bizerte lagoon was characterized by the highest concentrations of chromium (26.393 ppm) and manganese (281.748 ppm). While the highest copper content (39.098 ppm) was observed in El Bcherliya. The Korba lagoon revealed the highest concentration in cobalt and rubidium with 8.311 and 15.814 ppm, respectively. Bhiret El Biben was characterized by the highest concentration of lithium (29.087 ppm), strontium (2101.549 ppm) and tin (7.340 ppm). In addition, the lowest concentration for all the heavy metals studied was recorded in the bank of Sidi Ali Mekki lagoon. The different metals analysed in these lagoons do not exceed the maximum tolerate values [42] except lead that exceeds 100 ppm in the northern lagoon of Tunis (133.556 ppm).

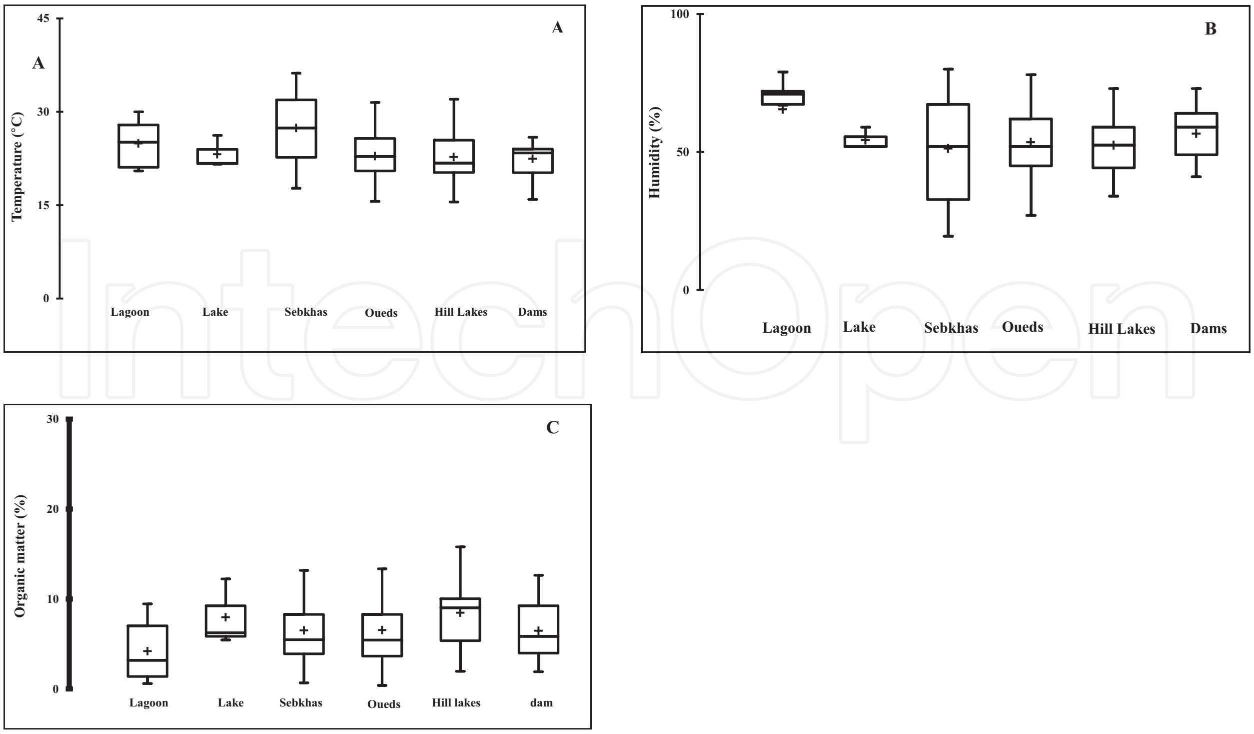

Figure 1. Environmental factors (A: Temperature (°C), B: Humidity (%), C: Organic matter (%)) measured at each wetland types.

In the second type of wetlands, lakes, the highest contents of manganese (1806 ppm), zinc (131.955 ppm), arsenic (4.211 ppm), cadmium (0.678 ppm), thallium (0.170 ppm) and lead (47.060) were observed in Ichkeul lake. Furthermore, Bouhnach lake was characterized by the important contents of lithium (7.735 ppm), vanadium (23.893 ppm), chromium (15.711 ppm), cobalt (7.497 ppm), nickel (16.567 ppm), strontium (643.783 ppm) and tin (0.140 ppm) and it was rather Majin Chitan lake that presented the highest concentration of copper (15.577 ppm) and rubidium (11.632 ppm). According to Henin [42], these stations were not contaminated since the content of heavy metals does not exceed the maximum tolerated values.

Concerning sebkhas, the highest concentrations in vanadium, rubidium and thallium were recorded in Sebkha Halk Menzel (36.357, 19.239 and 0.140 ppm, respectively). For chromium, manganese and tin, the highest concentrations were recorded in sebkhas of Sidi Khlifa (554.628 ppm), Moknine (387.880 ppm) and Sousse (2.793 ppm), respectively. The highest concentration of cobalt, nickel, strontium and cadmium was, respectively, observed in the Halk Menzel (10.460 ppm), Raoued (23.106 ppm), Sidi El Hani (3305.249 ppm) and Golla (0.422 ppm). Concerning copper, arsenic and lithium, the highest concentration was, respectively, recorded in sebkha Ariana (23.238 ppm), sebkha Golla (14.507 ppm) and sebkha Kairouan (41.861 ppm), and it was rather the sebkha Monastir that showed the highest concentration of zinc (86.453 ppm) and lead (48.741 ppm). According to these results and taking into account the tolerance thresholds, no sebkha was considered polluted with the exception of sebkha Sidi Khlifa that was considered as polluted by chromium which exceeds the maximum tolerated value (150 ppm) [42].

In wadis, the highest concentrations of vanadium (44.619 ppm), chromium (40.413 ppm), zinc (147.822 ppm) and lead (303.910 ppm) were recorded in Lahmam wadi. The highest concentration of cobalt (19,723 ppm), nickel (29,283 ppm), rubidium (27,016 ppm), and thallium (0.183 ppm) were found in Zit wadi. Whereas for lithium, manganese, copper, arsenic, strontium, cadmium and tin, the highest concentrations were, respectively, recorded in Soufi wadi (26.527 ppm), Mazbla wadi (644.069 ppm), Joumin wadi (19.448 ppm), Lanj wadi (9.380 ppm), Khniss wadi (1410.100 ppm), Bargou wadi (1.412 ppm) and El Korsi (1.019 ppm). According to Henin [42], the different prospected wadis were not polluted except for Joumin, Lassoued and Bargou wadis, which were considered as polluted with cadmium whose percentage exceeds the maximum tolerated value (0.7 ppm) as well as Lahmam wadi in which a lead concentration exceeded 100 ppm.

Sixteen hill lakes and 17 dams belonging to different bioclimatic stages were prospected. In the banks of these closed and artificial ecosystems, no amphipod was found.

Concerning heavy metals, in hill lakes, the highest concentrations of vanadium (46.795 ppm), cobalt (14.661 ppm), nickel (30.362 ppm), copper (16.611 ppm) and lead (23.047 ppm) were observed in Sidi Daoued hill lake. The highest concentration of zinc (88.804 ppm), arsenic (4.590 ppm), cadmium (3.031 ppm), thallium (0.176 ppm) was found in Zrab wadi hill lake. The khlifa wadi hill lake was characterized by the important content of lithium (24.891 ppm) and strontium (930.812 ppm) and it was rather Ouled Ali hill lake that presented the highest concentration of rubidium (16.526 ppm) and tin (0.629 ppm). Hill lakes of Khalsi and Ksayir Hamdoun were characterized by the highest concentration of chromium (43.394 ppm) and manganese (530.039 ppm). This analysis of heavy metals revealed that only Ain Ben Ali, Zad and Khalsi hills lakes were contaminated by the cadmium. Concerning dams, our results showed that Sidi Barrak dam was characterized by the highest concentration of majority of heavy metals, namely manganese (1060.291 ppm), cobalt (14.085 ppm), copper (22.840 ppm), zinc (151.90 ppm), arsenic (6.246 ppm), thallium (0.544 ppm) and lead (166.067 ppm); while Kasseb dam showed the highest concentration of vanadium (37.377 ppm), chromium (41.476 ppm), nickel (32.579 ppm) and rubidium (18.714 ppm). The highest concentration in lithium (15.013 ppm), cadmium, (5.426 ppm) and tin (0.137 ppm) was found in Gattar dam; while that of strontium (731.645 ppm) was observed in Jneyhiya dam.

3.3. Species richness

Eight species of amphipoda Talitridae, namely *Orchestia montagui* Audouin, 1826, *Orchestia mediterranea* Costa, 1853, *Orchestia gammarellus* (Pallas, 1766), *Orchestia stephensen*i Cecchini, 1928, *Orchestia cavimana* Heller, 1865, *Platorchestia platensis* (Kroyer, 1845), *Deshayesorchestia deshayesii* (Audouin, 1826) and *Talitrus saltator* (Montagu, 1808) were collected in different wetlands.

Species richness (S) varied between stations of the same as well as the different types of wetlands.

In lagoons, species richness varied between one species in El Bcherliya and eight species in the bank of Bizerte lagoon near Menzel Jmil. The differences observed between lagoons were highly significant ($F = 5.317$; $df = 9$; $p < 0.0001$). In the bank of lakes, amphipods were collected only in Ichkeul lake ($S = 5$).

Concerning sebkhas, among 30 sebkhas studied, talitrids were found in only four sebkhas namely: El Ouafi, Maâmoura, Moknine and Gargour. Species richness was equal to one species in the bank of sebkhas El Ouafi and Maâmoura namely *Orchestia gammarellus* and *Talitrus saltator*, respectively. In two other sebkhas, *Orchestia gammarellus* and *Orchestia mediterranea* were collected.

For wadis, individuals were collected only in six wadis among the 41 stations prospected. Species richness varies from one (El Fared wadi, Laakarit wadi, Khniss wadi and Lebna wadi) to six species (El Korsi). In hill lakes and dams, no species were collected.

3.4. Relative abundance and density

A total of 340 specimens of amphipoda Talitridae were collected in lagoons. The bank of Bizerte lagoon revealed statistically the most important relative abundance of amphipod community (36.04%) (Anova test: $F = 5.330$, $df = 9$, $p < 0.0001$). Moreover, in this station, *Orchestia mediterranea* was the most abundant species (25.7%). However, in the banks of El Bcherliya, the Ghar El Melh old harbour, Tunis north and south lagoons, it was rather *Orchestia gammarellus* that dominated. These two species have the same relative abundance (46.7%) in bank of Sidi Ali Mekki lagoon. In Bhiret El Biben lagoon, *Orchestia montagui* was the most abundant species (28.3%). The Anova test revealed that differences between the different lagoons were highly significant ($F = 7.922$; $df = 7$; $p < 0.0001$). The mean community density varied between 0.5 ind.m^{-2} in the bank of El Bcherliya and 241.5 ind.m^{-2} in that of Bizerte lagoon. Furthermore, our results showed that *Orchestia mediterranea* presented the most important density in the bank of Bizerte lagoon (62 ind.m^{-2}). Whereas, in the bank of El Bcherliya, Ghar El Melh old harbour, Tunis north and south lagoons, it was *Orchestia gammarellus* that exhibited the largest density with, respectively, 0.5, 19, 34 and 34.5 ind.m^{-2}. These two species were recorded with the same mean density in the bank of Sidi Ali Mekki lagoon (3.5 ind.m^{-2}). In lakes, 170 individuals were collected where *Orchestia mediterranea* presented the highest abundance (26.5%) and density (22.5 ind.m^{-2}).

In sebkhas, 352 specimens of amphipods were collected. Sebkha Gargour revealed the highest relative abundance (50%) followed by sebkha Moknine which abundance was equal to 34.66%. However, in the bank of the two other sebkhas, the abundance was relatively low in Mâamoura with 15.06% and very low in sebkha El Ouafi with 0.28%. The Anova test revealed a highly significant difference in relative abundance between these sebkhas ($F = 8.288$, $df = 29$, $p < 0.0001$). The relative abundance of *Orchestia gammarellus* and *Talitrus saltator* were maximal (100%), respectively, in sebkha El Ouafi and Mâamoura since each sebkha harbours only one species. In Moknine, abundance was greater for *Orchestia gammarellus* (53.3%) than in *Orchestia mediterranea* (46.7%) and inversely in the sebkha Gargour where the highest abundance was recorded for *Orchestia mediterranea* with 59.1%. In addition, no significant difference in relative abundance between species was found (Anova test: $F = 1.461$, $df = 2$, $p = 0.233$). The global mean density oscillated between 0.5 ind.m^{-2} in sebkha El Ouafi and 88 ind.m^{-2} in sebkha Gargour. The study of the mean density per species showed a very low density of

Orchestia gammarellus in sebkha Ouafi (0.5 ind.m^{-2}); this density became more pronounced in sebkha Moknine and Gargour with, respectively, 32.5 and 36 ind.m^{-2}. In these two last stations, *Orchestia mediterranea* had a density of 28.5 and 52 ind.m^{-2}, respectively.

Concerning wadis, 558 individuals were found. The most important global mean density was observed in the bank of El Korsi. *Orchestia mediterranea*, species living in allopatry in Khniss, Laakarit and El Fared wadis showed a density, respectively, equal to 49.5, 47.5 and 0.5 ind.m^{-2}; whereas, where it was in sympatry, its density was equal to 16 (El Korsi) and 7.5 ind.m^{-2} (Tinja). Furthermore, *Talitrus saltator*, which was the only amphipod collected in Lebna wadi estuary (67 ind.m^{-2}), showed a relatively lower density in El Korsi (13 ind.m^{-2}) and Tinja (6 ind.m^{-2}).

3.5. Diversity

According to the Simpson index (Is), the most important diversity was observed in the Bizerte lagoon where we noted the highest value which tends towards the specific richness (6.059) and the community was more balanced in Boughaz.

In Ghar El Melh old harbour, we obtained the lowest diversity compared to that observed in Boughaz. This result could be explained by the fact that this index does not consider rare species into account.

The Shannon-Weaver (H′) index ranged from 1.287 in the bank of Sidi Ali Mekki lagoon to 2.771 in the bank of Bizerte lagoon where the diversity was relatively significant. This index, which takes into account the rare species, was often accompanied by the equitability index, which was more or less insensitive to specific richness. It ranged from 0.812 (Sidi Ali Mekki) to 0.996 (Boughaz) where the community was more balanced.

In the banks of different sebkhas, we did not observed a great diversity, so the analysis of diversity was not carried out.

In wadis, results showed that the Simpson index varies between 1 in the Lebna wadi estuary, Khniss, Laakarit and El Fared wadis and 5.78 in El Korsi station where we found the highest species richness (S = 6). The Shannon-Weaver index confirmed the previous index showing that the most important diversity was observed in El Korsi station (H' = 2.56). Moreover, the study of the equitability index showed that the community was more balanced in this station (J' = 0.99) where species were equitably distributed.

3.6. Amphipod distribution according to environmental factors and wetland types

To better understand the species distribution in the different wetland types and to elucidate the parameters involved in their distribution, a canonical correspondence analysis was carried out (**Figure 2**). The first three axes, F1, F2 and F3 extract, respectively, 71.43, 20.23 and 5.55% of the variance. The two species, *Orchestia mediterranea* and *Orchestia gammarellus* that dominated the majority of lagoons and sebkhas were positively correlated with the strontium content and negatively with concentrations of vanadium, chromium, manganese, cobalt, nickel, copper, zinc, arsenic, rubidium, cadmium, thallium and lead. However,

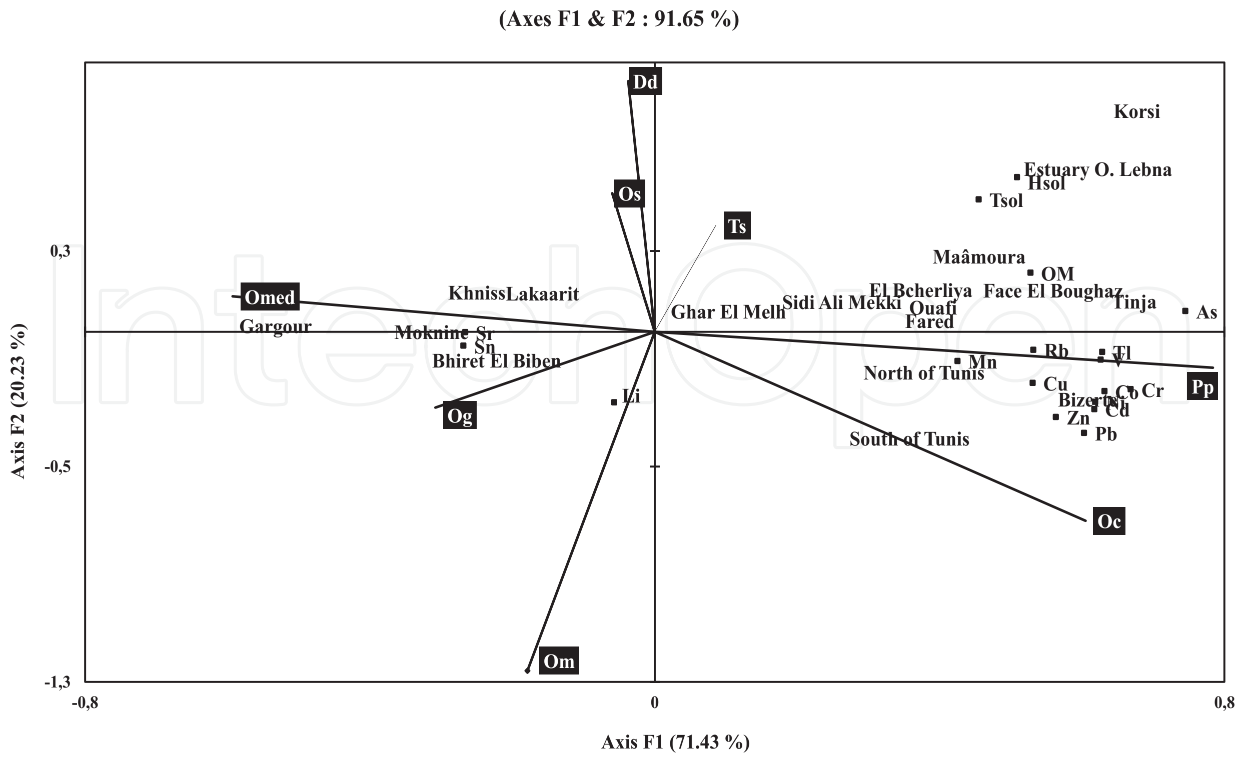

Figure 2. Canonical correspondence analysis (CCA) performed on the abiotic parameters. Om: *Orchestia montagui*, Og: *Orchestia gammarellus*, Omed: *Orchestia mediterranea*, Os: *Orchestia stephenseni*, Oc: *Orchestia cavimana*, Pp: *Platorchestia platensis*, Ts: *Talitrus saltator*, Dd: *Deshayesorchestia deshayesii*, Hsol: soil humidiy, Tsol: soil temperature, Gra: granulometry, V: vanadium, Cr: chromium, Mn: manganese, Co: cobalt, Ni: nickel, Cu: copper, Zn: zinc, As: arsenic, Cd: cadmium, Sn: tin, Tl: thallium, Pb: lead, Li: lithium, Rb: rubidium and Sr: strontium.

Talitrus saltator, abundant species in El Korsi and the estuary of Lebna wadi, as well as *Orchestia stephenseni* and *Deshayesorchestia deshayesii* were positively correlated with temperature and humidity. In the third axis, *Orchestia cavimana* and *Platorchestia platensis* were found positively correlated with organic matter and negatively with lithium and tin content.

4. Discussion

The study of the biodiversity talitrid populations in six types of wetlands revealed differences between these types.

The different prospections carried out in wetlands showed globally that the most important species richness was observed in lagoons. Moreover, no individual was collected in hill lakes and dams. Among lagoons studied, the bank of Bizerte lagoon was the most diverse one with eight species. This result was confirmed by several diversity indices performed in the present study.

Our hypothesis concerning the existence of a north/south diversity gradient was confirmed only for wadis. Furthermore, we did not reveal any significant difference concerning the vulnerability of lentic wetlands compared to the lotic type. Concerning Oniscidean group collected from the banks of Tunisian wetlands where many species were found in sympatry with amphipods, a positive correlation between species richness and altitudinal gradient has been highlighted [43]. The same authors showed that species richness differs significantly depending on wetland types or bioclimatic zones.

A total of 2420 amphipods belonging to different species were determined in all prospected wetlands; more than half of the specimens were collected in lagoons (N = 1340) with a mean density of 241.5 ind.m^{-2} observed in the bank of the Bizerte lagoon; *Orchestia mediterranea* showed the most important density in this lagoon. In sebkhas and wadis, the highest densities were recorded, respectively, in sebkha Gargour and El Korsi where *Orchestia mediterranea* and *Platorchestia platensis* were characterized by the highest density, respectively. Studying *Talitrus saltator* and *Britorchestia brito* populations in Zouara beach, Charfi-Cheikhrouha et al. [26] determined a mean density equal to 262.94 ± 85 ind.m^{-2}. These authors showed that the density increased in autumn and winter and reached a maximum in March for *Talitrus saltator*; while for *Britorchestia brito*, this density increased from the middle of March and peaked in October. In Algeria, in the bay of Bou Ismail, *Orchestia montagui* and *Deshayesorchestia deshayesii* reached more than 45,000 ind.m^{-2} [44]. In the Bou Regreg estuary, *Orchestia mediterranea* showed densities ranging from 3380 (February) to 7000 ind.m^{-2} (August) [45]. Studying the spatio-temporal distribution of amphipods in different wetlands in Tunisia, Jelassi [46] highlighted that the most important densities were observed during spring.

The diversity of the different talitrid species was related to the presence of different parameters. This relation depends on the wetland type; for example, in lagoons, the sandhopper *Talitrus saltator* was correlated with climatic (temperature, humidity) as well as edaphic factors (organic matter, granulometry, heavy metals of soil) whereas in sebkhas and wadis, this species was correlated only with edaphic factors. These results were also observed for other species. In this context, several studies have investigated the role of environmental factors and have revealed the influence of some factors rather than others. Jelassi et al. [31] have shown that talitrid abundance in the bank of Bizerte lagoon was closely related to air temperature. Bouslama et al. [47] showed that temperature was the important factor influencing the zonation whose augmentation induces the displacement or the migration of *Talitrus saltator* population from the top to the bottom of the beach. This result was similar to that found by Fallaci et al. [48], who indicated that mean zonation of this species was influenced by temperature during its activity period. Other authors such as Colombini et al. [49] confirmed the importance of sediment parameters in the selection of specific distribution area especially for young individuals.

Our results showed that the two species *Orchestia cavimana* and *Platorchestia platensis* were correlated with organic matter. Jelassi et al. [32] highlighted that air and soil temperature were the best predictors for *O. stephenseni* abundance that negatively corresponded with the proportion of fine sand fraction and organic matter content of the soil. *O. montagui* and *O. cavimana* abundances corresponded positively with air humidity and the soil lithium and rubidium contents, but negatively with the soil tin content and the proportion of the silt and clay fraction. *D. deshayesii* and *P. platensis* did not exhibit any clear correspondence with station characteristics.

According to Williams [50], the relationship between population movements and trophic preferences does not seem to be a major parameter in the structuring of zonation despite the important mobility of the sandhopper *Talitrus saltator*, which would induce a greater choice of nutrient sources. Studying the biodiversity of amphipods in some coastal lagoons in Tunisia, Jelassi et al. [33] showed that the most important species richness observed in the bank of Bizerte lagoon would be related to the presence of important vegetation in spring as well as the *Cymodocea nodosa* leaf litter and a high percentage of organic matter.

Attention was also given to biodiversity and biogeography for Oniscidean communities living in sympatry with amphipods in different wetland types prospected in the present study. Khemaissia et al. [43] showed that *Porcellio lamellatus, Tylos europaeus, Armadilloniscus ellipticus, Armadillo officinalis, Porcellio sexfasciatus* and *Chaetophiloscia elongata*, abundant species in the banks of lagoons, were associated with sodium content, pH and temperature of soil. However, other species such as *Armadillidium pelagicum, Armadillidium sulcatum, Armadillidium vulgare, Armadillidium boukornini, Armadillidium tunisiense* and *Porcellio dominici* were abundant around dams and hill reservoirs and were positively associated with elevation. The distribution of *Leptotrichus panzerii* and *Armadillidium granulatum*, in the sebkhas, was correlated with calcium content and humidity of soil.

Through these results, we did not reveal any significant difference regarding the vulnerability of lentic type wetlands compared to the lotic type wetlands. In order to test this hypothesis, the number of this last wetland type (lotic type) should be multiplied.

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Disclosure statement

No potential conflict of interest was reported by the authors.

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