

## Bio-indicator Potential in barbel *barbus callensis* (valenciennes, 1842) of the Seybouse basin (North- eastern, Algeria)]

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### Abstract

This work, carried on a four-year cycle (2011-2014) on habitats of different scales in the altitudinal Seybouse basin, aims to assess the bio-indicator potential in barbel (*Barbus callensis*, Valenciennes, 1842), rhéophile and endemic species, subservient to the running waters of the North African side. It attempts to examine the possible relationship between this species supposedly robust and its immediate environment at different scales in the altitudinal Seybouse basin. In this regard, a study was conducted on 21 selected physico-chemical parameters of 144 water samples, and 5 removed morphometric parameters on an overall sample of 443 specimens caught, from three stations belonging to three shorted sectors, although individualized by their physiographic and under anthropogenic pressures of different intensities. The results of the analysis of the collected data, confirmed through PCA, allowed us to identify a succession of two areas from upstream to downstream on the river system very clearly displaying a spatial segregation between size classes fish habitat (i) an upstream part on the tributaries formed by individuals with relatively good growth performance, which is characterized by low pollution and (ii) a downstream zone on the mainstream, formed by individuals of smaller size classes quite variable according to the habitat, which is characterized by a heavy pollution. The adult individuals are less resilient and prefer living waters and well oxygenated upstream and in the middle of the basin, not finding a suitable environment downstream, they were replaced by individuals of smaller size, less demanding and prefer relatively calmer waters.

### 1. Introduction

With a catchment area of approximately 6471 km<sup>2</sup>, Wadi Seybouse represents one of the most important rivers in Algeria [1]. In this watershed there are intense agricultural activities (cereals and market gardening) and industrial activities (more than 70 factories, the most important of them are in the vicinity of the maritime Seybouse) [2]. Its hydrographic network, made up of five dams, is home to an indigenous ichthyofauna (*Pseudophoxinus punicus*, *Anguilla anguilla*, *Barbus callensis*) [3] and Allochthonous (*Cyprinus carpio* and *Carassius carassius*) [4].

In Algeria, works on the systematic, biology and spatial distribution of continental fish populations is very old [5-12].

Over the past two decades, interest in the ecobiology of ichthyological stands of artificial and natural waterbodies has increased further due to commercial exploitation of this fishery resource in some localities in the country [13-16, 4, 17].

On the entire Seybouse basin, the riparian population exercises throughout the year an artisanal and sport fishing. In this ecosystem, Cyprinidae *Barbus callensis* (Valenciennes, 1842), an indigenous and endemic species, is found in abundance throughout the North African continental hydrographic network, and represents the main [7]. In addition, this species of high heritage value is listed on the IUCN (International Union for Conservation of Nature) Red List as a "minor concern" [18].

Because of its importance, The Seybouse has been the subject of several studies, both hydrological and morphodynamic [19, 20], hydrochemical [21-24] and ecological [25, 26].

The present work attempts to analyze the response of *B. callensis* subjected to various anthropogenic pressures (agricultural, domestic and industrial activities) on the scale of the Seybouse basin. The main objective is to assess the bioindicator potential of this species with a view to establishing an inventory in each river basin district and to identify the main anthropogenic pressures. However, the spatial dynamics of fish populations is an all the more relevant indicator of the overall quality of watercourses downstream of the trophic chain [27].

Apart from some brief investigations on the subject [28-30], Very little work is available on the response of fish populations to environmental pressures in response to an increasing demand from managers who often have to act in an increasingly harsh regulatory context [31].

Our study is in line with this problem and attempts to examine more precisely the relations between the barbel *B. callensis* population and its immediate environment at different altitudinal scales of the Seybouse basin.

## 2. Materials and Methods

### 2.1. Study environment

The study area in the Wadi Seybouse basin (area: 6471 km<sup>2</sup>, length: 240 km) is represented by three vast and distinct physiographic domains: The High Plains, the South Tellian and the Northern Tellian. It is the most extensive basin, after that of Medjerda, in the eastern part of North Africa [32]. The only important watercourse is the Wadi Seybouse, formed shortly before its entry into the commune of Guelma, Wadi Cherf and Wadi Bouhamdane. These two streams are main tributaries that flow into the Seybouse at the confluence upstream of the municipality of Medjez Amar.

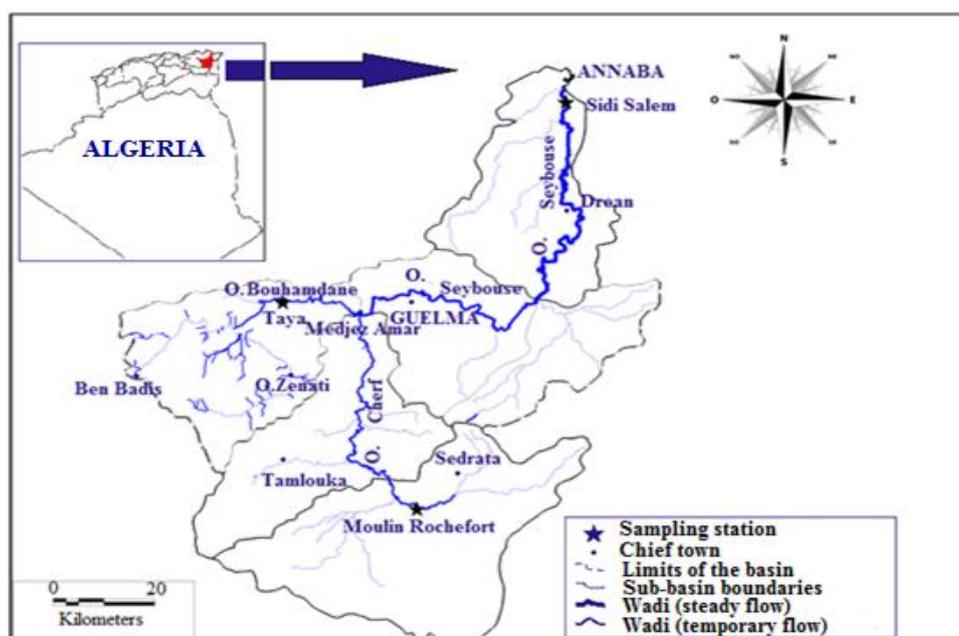
### 2.2. Choice of sampling stations

Given the size of the Seybouse basin, the study was carried out at three sites representative of the three hydrogeographic regions (Figure 1) taking into account the various activities identified in the area (agro-food industries, artisanal fisheries, domestic and agricultural waste) and defined as follows: (SI) the Moulin Rochefort as a moderately urbanized site with a productive and threatened ecosystem, (SII) the Taya as a site remarkable for its biodiversity and landscape quality, it is sheltered and is close to an intense agricultural activity area, (SIII) located at the mouth of Sidi Salem as a heavily anthropized site (Subject to urban, industrial and agricultural discharges), which requires opening-up initiatives. Each observation station was subjected to a GPS point and then easily positioned on the map of the watershed.

- **Station I** (Moulin Rochefort): (36 ° 03'34.56 "N - 007 ° 26'23.23" E), remote from anthropogenic activities, the station is located upstream of the basin on the Cherf (88, 61 km), tributary left bank of the Seybouse. In this zone, the whole of the relief is quite simple in its structure. The wadis and their tributaries converge jointly towards the Moulin Rochefort. This area extends over the plains of Sedrata and Tamlouka.

- **Station II** (Taya): (36 ° 28'00.76 "N - 007 ° 08'34.83" E), average course (average Seybouse), This area is located at an altitude of 378 m, in the South- Tellian region on the Bouhamdane (37,49 km) tributary left bank of the Seybouse. The basin of the average Seybouse is enclosed between the high plains in the south and the numidic chain in the North. This sector constitutes the essential part of the basin. It is drained by the Wadi Bouhamdane which takes its source at Bordj-Sabath and the downstream part of Wadi Cherf tributary of right bank of the Seybouse which descends of Moulin Rochefort. It is a stream with a rather sloping mountain profile with granulometry dominated by blocks. This environment, which is not influenced by anthropogenic activities, is located in forested area (hardwoods); the station is located within a relatively large area of agricultural influence.

- **Station III** (Sidi Salem): (36 ° 51'38.22 "N - 007 ° 46'04.57" E), this station is located in the northern Tellian area on the downstream of the main stream of the Seybouse (134, 74 Km), about 1 km from the mouth, almost at sea level (altitude: 2 to 3 m). It includes the Seybouse maritime, the Wadi Ressoul basin, Wadi Dardar and the littoral plains zone. The Basse Seybouse presents an asymmetrical relief with a slightly mountainous zone, relatively accentuated on the left bank of the wadi Seybouse. The sampling point is located downstream, of a strong agglomeration whose accumulation of domestic, industrial and agricultural pollution is remarkable.



**Figure 1:** Limits of the Seybouse basin and location of sampling stations.

### 2.3. Sampling plan

A total of 443 individuals were captured for monthly sampling campaigns. Sampling took place in the period from 2011 to 2014. Electrical fishing gear and trammel netting were used, the specimens were harvested as a first approach to electric fishing, with net fishery only as a last resort, if the first one fails or gives incomplete results, depending on the bathymetry, temperature and conductivity of the water body of the study site. The duration of immersion of the machine extends from 3 to 4 hours. At each station, it was also envisaged to take sufficient water to be analyzed in the laboratory. The extent of the sampling station has a fluvial surface whose length corresponds at least ten times the width of the wet bed, relying on four successive passages [27].

### 2.4. Environmental variables

For each of the 3 stations, the following physicochemical parameters: pH, temperature (T), dissolved oxygen (OD), electrical conductivity (in the case of 197i WTW) EC, salinity (SAL) and reduction potential (Eh). Turbidity (TU), expressed directly in NTU, was estimated using a HACH 2100 N turbidimeter. The suspended materials (SM), expressed in mg/l, were obtained by differential weighing of the filter before and after filtration after drying at 105 °C. On a Millipore filtration ramp using Whatman GF/C filters of 47 µm of porosity.

The dry residue (RS) was obtained by weighing a certain volume of water evaporated at 110 °C, in a calibrated dish. In addition to the in situ measurements, different minerals and nutrients were analyzed separately in each station using water samples taken at 3 m from the bank and at an average depth of 40 cm.

The samples were placed in hermetic plastic bottles and transported to the laboratory in a cooler maintained at 4 °C for immediate processing. The physicochemical analyzes, expressed in mg/l: ( $\text{NO}_3^-$ ,  $\text{NO}_2^-$ ,  $\text{PO}_4^{3-}$ ,  $\text{NH}_4^+$ ,  $\text{SO}_4^{2-}$ ,  $\text{Ca}^{2+}$ ,  $\text{Mg}^{2+}$ ,  $\text{K}^+$ ,  $\text{CaCO}_3$  et  $\text{Cl}^-$ ) With the exception of TH ( $^0\text{f}$ ), were carried out in the laboratory using a multiparameter photometer (Hanna Instruments HI 83200, Romania).

### 2.5. Fishing techniques and biometrics

Parallel to the measurements of the physicochemical parameters, fish sampling campaigns were conducted with the same frequency on the habitats of the stations delimited for this study. Depending on the bathymetry of the station, fish trapping occurred with two distinct but complementary fishing gears. The trammel net (L = 20 m, core mesh void = 95 mm and bundle void = 165 mm) was used at depths greater than 1 m, while the portable DC voltage generator (Between 500 and 800 V) was only used in shallow waters (<0, 5 m), upstream the watercourse until the water became clear in the vicinity of the experimenter. The electric field generated around the anode is active over an area of about 1.5 to 2 m<sup>2</sup> which constitutes the so-called "attractive" or "efficient" zone. All fish in the adjacent area will react to this electrical current.

The product of each capture is transported to the laboratory in a cooler at 4 °C to be sorted and identified up to the species [33-35, 10]. For each species, we carried out the following biometric measurements:

(i) the total length (Lt) is measured from the tip of the muzzle (but excluding the barbs) to that of the caudal fin whose lobes are brought together to give a maximum value to the measure [36] ;

(ii) the fork length (Lf) is also measured from the tip of the muzzle to the median notch and;

(iii) the standard length (Ls) measured from the tip of the muzzle to the tip of the hypural bone, or the fleshy part of the caudal peduncle, that is to say, excluding the caudal fin.

Once the measurements have been taken, the fish are then weighed (total weight: Wt and eviscerated: We) using a Sartorius electronic scale (accuracy:  $\pm 0,01g$ ). Only the species *B. callensis* was considered in this study.

## 2.6. Statistical analysis

In order to characterize the adaptive strategy of the *B. callensis* population to environmental conditions, a Principal Component Analysis (PCA) was performed. The intermediate correlation matrices, the correlation coefficients between the variables and the two axes F1 and F2 and the projection of the variables in the space of the axes F1 and F2 were also obtained using the statistical analysis and processing software Statbox Of the data, version 2.5 for Windows. This method of analysis is used to understand the intensity of the relationships uniting a faunistic unit with the environmental factors [37].

Physico-chemical monitoring of the water will make it possible to establish the degree of pollution by comparing the concentrations of pollutants with quality thresholds (= environmental quality standards), according to the new grid of the system of assessment of water quality of watercourses SEQ-Water [38], which derives from the application of the European framework directive.

Finally, the comparison of the averages was carried out using the Tukey HSD test provided by the Minitab 17 software. The test was applied to a probability level of  $p = 0,05$  in order to investigate any significant differences between the averages. The 5% risk we have chosen is used to determine the critical value  $q$ , which is compared to the standardized difference between the averages.

## 3. Results

### 3.1. Physico-chemical characteristics of waters

Table 1 summarizes the averages of all physico-chemical parameters measured during the study period. The mean annual temperatures recorded at the three stations (SI = 16, 55 °C, SII = 15, 60 °C, SIII = 18, 13 °C) show no significant difference ( $p = 0,138$ ). The mean pH changed significantly from 7, 98 (SI) to 8, 86 in site III with a median value of 8.17 in station II ( $p = 0,001$ ). The mean electrical conductivity is different between the three stations ( $p = 0,000$ ); it is between 1151, 31  $\mu S/cm$  (SII) and 1598, 81  $\mu S/cm$  (SI). On the other hand, a value of 2623, 38  $\mu S/cm$  is recorded in station III, close to the mouth. Salinity also showed variable levels between the three stations ( $p = 0,001$ ) with maximum downstream values of 1, 26 mg/l (SIII). On the other hand, the average haline concentrations recorded in stations I and II are of the order of 0, 47 mg/l and 0, 32 mg/l, respectively.

The average dissolved oxygen content also tends to decrease significantly from upstream to downstream, with respective values of 5,34 mg/l at sites I and II and 4,80 mg/l at site III ( $p = 0,001$ ).

Mean SM concentrations showed no significant difference ( $p = 0,053$ ) between the studied sites (SI = 27, 70 mg/l, SII = 27, 54 mg/l, SIII = 26, 96 mg/l), as is the case with surface water turbidity ( $p = 0,251$ ) where the mean values recorded are 60, 30 NTU (SI), 128, 99 NTU (SII) and 43, 25 NTU (SIII), respectively.

The proportional relationship of the dry residues (DR) of this parameter with the electrical conductivity (multiplicative factor between 0, 75 and 0, 8) is possible, so this parameter is not necessary to be retained here as all Eh,  $K^+$  and OM, in the absence of quality class thresholds.

The recorded averages of calcium carbonate ( $CaCO_3$ ) in the three stations are generally less than 200 mg/l. They increased progressively from 165, 78 mg/l (SII) to 192, 75 mg/l (SIII) to a maximum of 213, 19 mg/l in station I ( $p = 0,087$ ).

The hardness (TH) of the water varies significantly between the three stations ( $p = 0,005$ ); the mean values are higher in station I (70, 60 °f) than in stations II (54, 10 °f) and III (65, 53 °f).

The mean chloride content varies significantly between the three stations ( $p = 0,000$ ). The highest value was recorded in station III (960.40 mg / l) downstream of the basin, compared with relatively lower values in stations I (230,10 mg/l) and II (191,27 mg/l).

Mean nitrate concentrations fluctuated significantly between 27,53 mg/l and 26,95 mg/l, respectively for stations I and III to 36,79 mg/l in station II ( $p = 0,805$ ), such as nitrite (SI = 0,90 mg/l, SII = 0,94 mg/l, SIII = 3,51 mg/l) ( $p = 0,000$ ), of ortho-phosphates (SI = 10,86 mg/l, SII = 17.58 mg/l, SIII = 26.42 mg/l) ( $p = 0,000$ ), ammonium (SI = 3,46 mg/l, SII = 1,32 mg/l et SIII = 3,23 mg/l) ( $p = 0,000$ ), sulphates (SI = 77,31mg/l, SII = 94,63mg/l, SIII = 309,93mg/l) ( $P = 0,000$ ) calcium (SI = 135,45 mg/l, SII = 72.23 mg/l SIII = 111,55 mg/l) ( $p = 0,001$ ) and magnesium (SI = 81,78 mg/l; SII = 61,86 mg/l; SIII = 86,22 mg/l) ( $p = 0,026$ ).

**Table 1:** Annual mean values of physico-chemical analyzes in the three stations surveyed during the sampling period (2011-2014)

Settings	Unit	Symbol	S I	S II	S III	Standards
Température	°C	T	16,55±1,10	15,60±1,03	18,93±1,26	20 à 25
pH	pH	pH	7,98±0,07	8,17±0,09	8,86±0,72	6,5 à 9,5
Electrical Conductivity	µS/cm	EC	1598,81±280,51	1151,31±60,30	2623,38±1303,32	180 à 3500
Salinity	g/l	SAL	0,47±0,19	0,32±0,04	1,26±0,74	1,00
Potentiel redox	mV	Eh	-108,13±23,16	-95,06±9,89	-103,25±37,56	-
Dissolved oxygen	mg/l	DO	5,34±0,61	5,34±0,09	4,80±0,36	8 à 4
Suspended matter	mg/l	SM	27,70±0,96	27,54±0,40	26,96±1,21	2 à 38
Turbidity	NTU	Turb	60,30±25,14	128,99±142,88	43,25±32,51	1à 70
Dry Residue	mg/l	DR	716,41±158,49	797,45±260,98	1998,36±936,52	-
Calcium carbonate	mg/l	CaCO <sub>3</sub>	213,19±5,52	165,78±3,34	192,75±4,31	< 200
Total Hardness	°f	TH	70,60±0,47	54,10±1,00	65,53±2,45	4 à 8
Chlorides	mg/l	Cl <sup>-</sup>	230,10±37,06	191,27±9,07	960,40±412,34	50 à150
Organic matter	mg/l	OM	3,52±0,08	3,30±0,05	6,65±0,41	-
Nitrates	mg/l	NO <sub>3</sub> <sup>-</sup>	27,53±0,66	36,79±1,70	26,95±0,86	2 à 25
Nitrites	mg/l	NO <sub>2</sub> <sup>-</sup>	0,90±0,07	0,94±0,07	3,51±3,31	0,03 à 0,5
Ortho phosphates	mg/l	PO <sub>4</sub> <sup>3-</sup>	10,86±15,26	17,58±29,12	26,42±43,58	0,1à 1
Ammonia	mg/l	NH <sub>4</sub> <sup>+</sup>	3,46±0,19	1,32±0,09	3,23±0,22	0,1 à 2
Sulphates	mg/l	SO <sub>4</sub> <sup>-</sup>	77,31±2,15	94,63±1,70	309,93±89,23	60 à190
Calcium	mg/l	Ca <sup>2+</sup>	135,45±1,62	72,23±0,87	111,55±6,68	32 à 300
Magnesium	mg/l	Mg <sup>2+</sup>	81,78±2,22	61,86±0,59	86,22±15,82	50 à 100
Potassium	mg/l	K <sup>+</sup>	35,38±1,25	53,13±0,78	71,56±17,26	-

### 3.2. Biometrics

A total of 443 specimens were captured. Table 2 summarizes the limit and mean metric and weight characteristics of *B. callensis* sampled at the three stations in the Seybouse Basin between 2011 and 2014. This sample is distributed a long a metric and weight gradient as follows:

- Station I: 177 individuals (387 < Lt <453 mm; 578 < We <978 g),
- Station II: 165 individuals (307 < Lt <434 mm, 296 < We <395 g) and,
- Station III: 101 individuals (176 < Lt <188 mm, 57 < We <73 g).

### 3.3. Statistical Comparison of Biotic and Environmental Characteristics

The results of the analysis of variance (ANOVA) using the Tukey HSD multiple comparison test of the mean metric, weight and physico-chemical values of the three physiographic domains taken in pairs are shown in Table 3. Analysis of the recorded data shows that in the three studied stations compared two to two ( $p < 0.05$ ), the mean difference is not significant for the parameters T<sup>o</sup>, Eh, SM, Turb, NO<sub>3</sub><sup>-</sup> and CaCO<sub>3</sub> but significant for the parameters pH, EC, SAL, DO, DR, NO<sub>2</sub><sup>-</sup>, SO<sub>4</sub><sup>-</sup>, NH<sub>4</sub><sup>+</sup>, Ca<sup>2+</sup>, Mg<sup>2+</sup>, TH, Cl<sup>-</sup>, K<sup>+</sup>, PO<sub>4</sub><sup>3-</sup> and OM. The same is true for morphometric descriptors where the comparison of the means and variances of the biometric characters using the Tukey test, taken two by two on the samples of the three sites, shows heterogeneity of the three fractions of populations of *B Callensis*.

**Table 2 :** Metric (mm) and weight (g) characteristics of *Barbus callensis* in the Seybouse Basin at the three stations surveyed during the sampling period (2011-2014).

Parameters		SI (N = 177)	SII (N = 165)	SIII (N = 101)
Total length (Lt)	Limit values	387.83 - 453.40	307.93 - 334.79	176.80 - 188.95
	Average ± S. Deviation	(412,59±28,34)	(319,22±11,50)	(183,79±05,08)
Fork length (Lf)	Limit values	352,32 - 412,02	276,46 - 298,33	157,97 - 168,26
	Average ± S. Deviation	(374,67±25,86)	(285,62±09,79)	(163,43±04,24)
Standard length (Ls)	Limit values	325,94 - 391,51	254,99 - 275,00	145,26 - 154,06
	Average ± S. Deviation	(349,35±28,94)	(263,64±8,64)	(150,11±03,64)
Total weight (Wt)	Limit values	632,79 - 140,77	348,18 - 444,31	066,41 - 084,04
	Average ± S. Deviation	(815,33±224,05)	(389,53±40,02)	(074,10±07,40)
Eviscerated weight (We)	Limit values	578,60 - 978,12	296,15 - 395,05	057,33 - 073,15
	Average ± S. Deviation	(725,53±174,80)	(334,19±42,52)	(064,09±06,61)

**Table 3:** Results of the analysis of variance (ANOVA), Tukey HSD multiple comparison test of metric and physicochemical averages taken two by two, recorded in the three stations surveyed during the sampling period (2011-2014).

Settings	S I	S II	S III	DL	Value of F	Value of p	Group		
							S I	S II	S III
Lt	412,59±15,06	319,22±18,07	184,77±6,66	2	263,34	0,000	A	B	AB
Lf	374,67±9,21	285,62±17,94	163,74±5,58	2	307,37	0,000	A	B	AB
Ls	349,35±12,23	263,64±17,38	150,40±5,66	2	247,07	0,000	A	B	AB
Wt	815,3 ±90,4	389,5±53,9	74,37±11,71	2	147,97	0,000	A	B	AB
We	725,5±65,6	334,2±57,5	64,30±8,94	2	172,33	0,000	A	B	AB
T °	16,96±3,60	17,09±4,27	20,39±5,77	2	2,10	0,138	A	A	A
pH	8,08±0,21	8,09±0,14	8,53±0,44	2	9,18	0,001	B	B	A
EC	1531±464	1177±231,5	2644±900	2	19,56	0,000	B	B	A
SAL	0,49±0,11	0,33±0,12	1,29±0,47	2	38,47	0,001	B	B	A
Eh	-94,21±31,25	-99,44±21,36	-90,29±28,	2	0,34	0,717	A	A	A
DO	5,42±0,29	5,53±0,39	4,95±0,42	2	8,30	0,001	A	A	B
SM	30,07±12,05	32,18±13,73	19,92±12,21	2	3,21	0,053	A	A	A
Turb	50,1±46,2	72,4±90,5	30,24±27,30	2	1,44	0,251	A	A	A
DR	786±240,7	1033±491,00	1734±731,00	2	10,43	0,000	B	B	A
CaCO3	221,8±49,8	186,3±60,9	174,1±46,9	2	2,64	0,087	A	A	A
TH	64,86±19,46	49,55±12,02	70,25±12,08	2	6,20	0,005	A	B	A
Cl <sup>-</sup>	217,2±100,7	194,8±48,0	1018,00±845	2	10,88	0,000	B	B	A
OM	3,54±1,53	3,07±1,62	6,30±1,56	2	14,78	0,000	B	B	A
NO <sub>3</sub> <sup>-</sup>	29,27±6,92	30,18±19,41	33,52±19,96	2	0,22	0,805	A	A	A
NO <sub>2</sub> <sup>-</sup>	0,75±0,45	0,87±0,55	13,20±9,08	2	22,20	0,000	B	B	A
PO <sub>4</sub> <sup>3-</sup>	12,89±3,72	16,31±4,70	32,82±15,43	2	14,92	0,000	B	B	A
NH <sub>4</sub> <sup>+</sup>	3,54±1,82	0,73±0,74	2,93±1,38	2	13,73	0,000	A	B	A
SO <sub>4</sub> <sup>-</sup>	107,8±44,9	121,6±47,5	276,2±141,8	2	12,92	0,000	B	B	A
Ca <sup>2+</sup>	143,32±31,72	97,08±27,57	116,60±20,55	2	8,86	0,001	A	B	AB
Mg <sup>2+</sup>	70,50±19,43	52,86±19,13	79,02±28,80	2	4,07	0,026	AB	B	A
K <sup>+</sup>	41,69±12,58	45,21±21,16	70,17±13,87	2	10,87	0,000	B	B	A

Averages sharing no letters are significantly different.

### 3.4. Principal Component Analysis (PCA) between Physicochemical and Biometric Characteristics

The first two factorial axes contribute respectively to 69.46% and 30.51% of inertia, a cumulative percentage of 99.97%. This result can be considered satisfactory given the relatively high number of variables processed (Table 4).

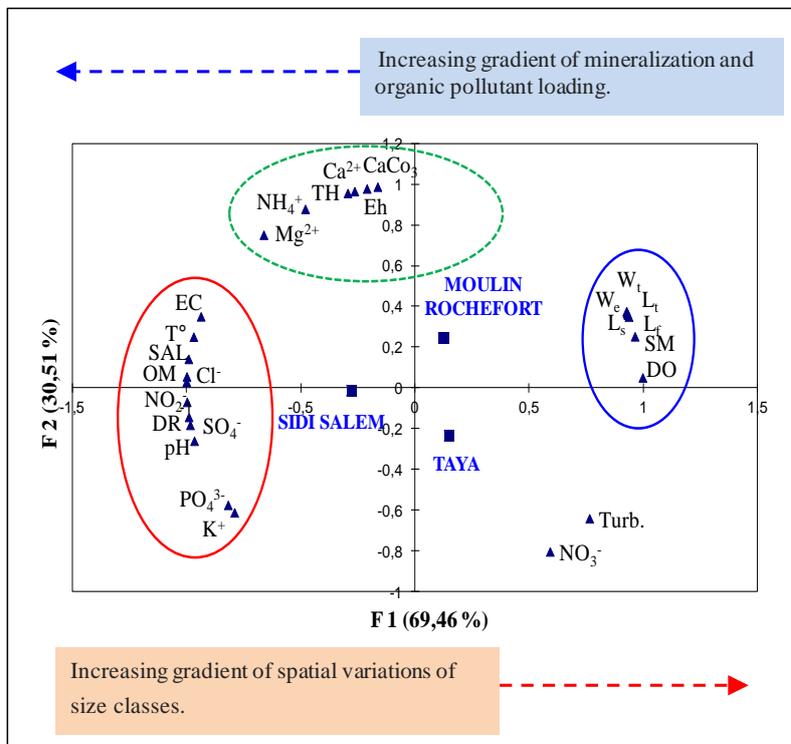
**Table 4:** Discrimination of groups of variables according to their coordinates (COOR) and their Relative contributions (CTR) on the first two principal components and their proximity, During the sampling period (2011- 2014).

Variables	Axis F <sub>1</sub> 69,46%		Axis F <sub>2</sub> 30,51%	
	COOR F <sub>1</sub>	CTR F <sub>1</sub>	COOR F <sub>2</sub>	CTR F <sub>2</sub>
Lt	0,93	0,87	0,35	0,12
Lf	0,92	0,86	0,36	0,13
Ls	0,92	0,86	0,37	0,13
Wt	0,93	0,87	0,34	0,12
We	0,93	0,86	0,36	0,13
T°	-0,96	0,93	0,24	0,06
pH	-0,96	0,93	-0,26	0,06
EC	-0,93	0,87	0,34	0,12
SAL	-0,99	0,98	0,14	0,01
Eh	-0,20	0,04	0,97	0,95
DO	0,99	0,99	0,04	0,00
SM	0,96	0,93	0,25	0,06
Turb	0,76	0,58	-0,64	0,41
DR	-0,98	0,97	-0,14	0,02
NO <sub>3</sub> <sup>-</sup>	0,59	0,35	-0,80	0,64
NO <sub>2</sub> <sup>-</sup>	-0,99	0,99	-0,07	0,00
SO <sub>4</sub> <sup>-</sup>	-0,98	0,96	-0,18	0,03
NH <sub>4</sub> <sup>+</sup>	-0,47	0,23	0,87	0,77
Ca <sup>2+</sup>	-0,26	0,06	0,96	0,93
Mg <sup>2+</sup>	-0,66	0,43	0,75	0,56
TH	-0,29	0,08	0,95	0,91
Cl <sup>-</sup>	-0,99	0,99	0,05	0,00
K <sup>+</sup>	-0,78	0,62	-0,61	0,37
PO <sub>4</sub> <sup>3-</sup>	-0,81	0,66	-0,57	0,33
CaCO <sub>3</sub>	-0,16	0,02	0,98	0,97
OM	-0,99	0,99	0,02	0,00

On the bipolar F1 axis, extracting more than 69,46% of the total variance, a maximum of variables is projected. At its positive pole (Figure 2), defined by the physicochemical parameters, the DO content (CTR = 0,99) and the SM (CTR = 0,93) are located. The same applies to morphometric parameters Lt (CTR= 0,87), Wt (CTR= 0,87), Lf (CTR= 0,86), Ls (CTR= 0,86) et We (CTR= 0,86) de *B. callensis* and turbidity (CTR = 0,58). On the negative side of the same figure, most of the variables are projected, such as the organic matter content OM (CTR = 0,99), NO<sub>2</sub><sup>-</sup> (CTR = 0,99), Cl<sup>-</sup>, CTR = 0,98), SO<sub>4</sub><sup>-</sup> (CTR = 0,96), pH of the water (CTR = 0,93), T° (CTR = 0,93), EC (CTR = 0,87), the water content of PO<sub>4</sub><sup>3-</sup> (CTR = 0,66) and K<sup>+</sup> (CTR = 0,62). The sector that contributed the most to the inertia of axis 1, negative side is Sidi Salem, up to 66.50%.

The F2 axis, which represents only 30,51% of the variance, at its positive pole project the water content in  $\text{CaCO}_3$  (CTR = 0,97), Eh (CTR = 0,95),  $\text{Ca}^{2+}$  (CTR = 0,93), TH (CTR = 0,91), water content of  $\text{NH}_4^+$  (CTR = 0,77) and  $\text{Mg}^{2+}$  (CTR = 0,56).

The station that contributed the most to the inertia of axis F2, positive side is Moulin Rochefort (SI), at 52,76%. At its negative pole the  $\text{NO}_3^-$  (CTR = 0,64) is projected. The station that contributed the most to the inertia of axis 2, negative side is Taya (SII), at 47,06%.



**Figure 2:** Projected variables and observations (sampling stations) in the basin Of the Seybouse during the sampling years on the F1 axes (in continuous lines) And F2 (dashed lines).

### 3.5. Relationship between the first two main components and the variables.

The two axes taken into account to describe the correlations between variables related to spatial structures, alone account for 99,97% of the total information with respectively 69,46% for axis F1 and 30,51% Axis F2.

In the plane formed by main components 1 and 2 (Figure 2), the variables most correlated to CP 1 are DO, SM, Lt, Wt, Lf, Ls, We, OM,  $\text{NO}_2^-$ ,  $\text{Cl}^-$ , SAL, DR,  $\text{SO}_4^-$ , pH,  $\text{T}^\circ$ , EC,  $\text{PO}_4^{3-}$  and  $\text{K}^+$ .

Among these variables, those that contributed most to the CP1, accounting for 69.46% of the variance, distinguish two different groups: the first grouping DO, SM, Lt, Wt, Lf, Ls and We, which are strongly correlated between And positively to F1, since they define eigenvectors of the same direction. The second group which is collected by the elements OM,  $\text{NO}_2^-$ ,  $\text{Cl}^-$ , SAL, DR,  $\text{SO}_4^-$ , pH,  $\text{T}^\circ$ , EC,  $\text{PO}_4^{3-}$  and  $\text{K}^+$ , which are negatively correlated to CP1.

On the axis defined by CP2, which represents only 30.51% of the variance, the variables most correlated to this main component are  $\text{CaCO}_3$ , Eh,  $\text{Ca}^{2+}$ , TH,  $\text{NH}_4^+$  and  $\text{Mg}^{2+}$ .

Other variables ( $\text{NO}_3^-$  and Turb) being far from the ends of the axis defined by CP1 and CP2 respectively, their correlation is certainly not very strong. These variables are probably better explained by other major components, other than CP1 and CP2.

### 3.6. Correlation matrix

Examination of the matrix of correlation between variables taken in pairs (Table 5) reveals the presence of a first set of variables, consisting of morphometric descriptors strongly correlated with each other (Lt/Lf, Lt/Ls, Lt/Wt, Lt/We, Lf/Ls, Lf/Wt, Lf/We, Ls/Wt, Ls/We and Wt/We), positive linear correlations since they define eigenvectors of the same direction ( $r > 0.99$ ,  $p < 0.05$ ). It is also noted that the different physicochemical

parameters correlate positively with each other (Eh/Ca<sup>2+</sup>, Eh/CaCO<sub>3</sub>, DR/NO<sub>2</sub><sup>-</sup>, DR/SO<sub>4</sub><sup>-</sup>, Ca<sup>2+</sup>/TH, Cl<sup>-</sup>/OM, K<sup>+</sup>/PO<sub>4</sub><sup>3-</sup>), In a very significant way (r > 0.99, p < 0.05). Moreover, the group of physicochemical parameters (pH/SM, DO/NO<sub>2</sub><sup>-</sup>, DO/OM, SM/SO<sub>4</sub><sup>-</sup>), correlate negatively with one another very significantly (r > 0.99, p < 0.05).

Table 5: Correlation matrix (Pearson (n)) between the variables studied.

	Lt	Lf	Ls	Wt	We	T°	pH	EC	SAL	Eh	DO	SM	Turb	DR	NO <sub>3</sub> <sup>-</sup>	NO <sub>2</sub> <sup>-</sup>	SO <sub>4</sub> <sup>-</sup>	NH <sub>4</sub> <sup>+</sup>	Ca <sup>2+</sup>	Mg <sup>2+</sup>	TH	Cl <sup>-</sup>	K <sup>+</sup>	PO <sub>4</sub> <sup>3-</sup>	CaCO <sub>3</sub>	OM	
Lt	1,00																										
Lf	<b>0,99*</b>	1,00																									
Ls	<b>0,99*</b>	<b>0,99*</b>	1,00																								
Wt	<b>0,99*</b>	<b>0,99*</b>	<b>0,99*</b>	1,00																							
We	<b>0,99*</b>	<b>0,99*</b>	<b>0,99*</b>	<b>0,99*</b>	1,00																						
T°	-0,81	-0,80	-0,80	-0,82	-0,81	1,00																					
pH	-0,99	-0,99	-0,99	-0,99	-0,99	0,86	1,00																				
EC	-0,74	-0,74	-0,73	-0,75	-0,74	0,99	0,81	1,00																			
SAL	-0,87	-0,86	-0,86	-0,87	-0,87	0,99	0,91	0,97	1,00																		
Eh	0,15	0,16	0,17	0,14	0,15	0,44	-0,05	0,53	0,34	1,00																	
DO	0,95	0,94	0,94	0,95	0,94	-0,95	-0,97	-0,91	-0,98	-0,16	1,00																
SM	0,99	0,98	0,98	0,99	0,99	-0,87	<b>-0,99*</b>	-0,81	-0,91	0,04	0,97	1,00															
Turb	0,48	0,47	0,46	0,49	0,48	-0,90	-0,57	-0,94	-0,84	-0,78	0,73	0,57	1,00														
DR	-0,97	-0,97	-0,97	-0,97	-0,97	0,92	0,99	0,87	0,95	0,06	-0,99	-0,99	-0,66	1,00													
NO <sub>3</sub> <sup>-</sup>	0,26	0,25	0,24	0,27	0,26	-0,77	-0,36	-0,83	-0,69	-0,91	0,55	0,37	0,97	-0,47	1,00												
NO <sub>2</sub> <sup>-</sup>	-0,95	-0,95	-0,95	-0,95	-0,95	0,94	0,98	0,90	0,97	0,14	<b>-0,99*</b>	-0,97	-0,71	<b>0,99*</b>	-0,53	1,00											
SO <sub>4</sub> <sup>-</sup>	-0,98	-0,98	-0,98	-0,98	-0,98	0,90	0,99	0,85	0,94	0,02	-0,99	<b>-0,99*</b>	-0,63	<b>0,99*</b>	-0,43	0,99	1,00										
NH <sub>4</sub> <sup>+</sup>	-0,13	-0,12	-0,11	-0,14	-0,13	0,68	0,23	0,75	0,59	0,95	-0,43	-0,24	-0,93	0,34	-0,99	0,41	0,31	1,00									
Ca <sup>2+</sup>	0,09	0,11	0,11	0,08	0,10	0,49	0,00	0,58	0,39	<b>0,99*</b>	-0,21	-0,01	-0,82	0,12	-0,93	0,19	0,08	0,97	1,00								
Mg <sup>2+</sup>	-0,34	-0,33	-0,33	-0,35	-0,34	0,82	0,44	0,88	0,75	0,87	-0,62	-0,44	-0,98	0,54	-0,99	0,60	0,51	0,97	0,89	1,00							
TH	0,06	0,07	0,08	0,05	0,07	0,52	0,03	0,60	0,42	0,99	-0,24	-0,04	-0,83	0,15	-0,94	0,22	0,11	0,97	<b>0,99*</b>	0,91	1,00						
Cl <sup>-</sup>	-0,91	-0,90	-0,90	-0,91	-0,91	0,98	0,94	0,95	0,99	0,26	-0,99	-0,94	-0,80	0,98	-0,63	0,99	0,97	0,52	0,31	0,70	0,34	1,00					
K <sup>+</sup>	-0,95	-0,96	-0,96	-0,95	-0,95	0,61	0,92	0,52	0,69	-0,43	-0,81	-0,91	-0,21	0,87	0,02	0,83	0,88	-0,15	-0,38	0,06	-0,35	0,75	1,00				
PO <sub>4</sub> <sup>3-</sup>	-0,96	-0,97	-0,97	-0,96	-0,97	0,64	0,93	0,56	0,72	-0,39	-0,84	-0,93	-0,25	0,89	-0,01	0,85	0,90	-0,11	-0,34	0,10	-0,31	0,78	<b>0,99*</b>	1,00			
CaCO <sub>3</sub>	0,20	0,21	0,21	0,19	0,20	0,40	-0,10	0,49	0,29	<b>0,99*</b>	-0,11	0,08	-0,75	0,01	-0,89	0,09	-0,02	0,94	0,99	0,84	0,99	0,21	-0,47	-0,43	1,00		
OM	-0,92	-0,91	-0,91	-0,92	-0,92	0,97	0,95	0,94	0,99	0,23	<b>-0,99*</b>	-0,95	-0,78	0,98	-0,61	0,99	0,97	0,50	0,29	0,68	0,32	<b>0,99*</b>	0,77	0,80	0,19	1,00	

The values in (\*) are significantly different from 0 to a significance level p < 0.05.

#### 4. Discussion

The mean values of the physicochemical parameters of the water show that the mean values of the water temperature have an increasing gradient from upstream to downstream, with a slight difference recorded at station II. These discrepancies can be due to the daily time difference between the different sampling points. Indeed, site II is relatively shaded and preserved in a forest context. Fluctuations in the temperature of a watercourse remain linked, on the one hand, to local conditions such as regional climate, topography, duration of sunshine, and on the other hand to flow and depth [39]. Obviously,, water temperature is an ecological factor that generates significant ecological impacts [40]. In general, Cyprinidae prefer warm, current, or stagnant, less mineralized waters [41] where adult individuals are often less tolerant of high temperatures than juveniles. This species is described as opportunistic with a capacity to adapt to temperature conditions between 36 and 40 °C [7].

From the point of view of electrical conductivity, the recorded values make it possible to classify the Seybouse in the class of water bodies in very good condition with respect to the mineralization. The high value of the electrical conductivity recorded in the vicinity of the downstream station (SIII) is closely related to the marine influence (Annaba gulf) and to the water inputs of the highly mineralized Wadi Meboudja (EC > 2100 µS / cm ), And which is fed by the salt waters of Lake Fetzara [42].

The increasing gradient of salinity observed from upstream to downstream of the Seybouse basin is linked either to intake of chlorides of anthropogenic origin or to natural salinization of water.

The region is directly subject to marine intrusions favored by its low inclined bed, which favors the rise of marine waters over long distances, especially during periods of storms where the values exceed the critical threshold of 1 g/l. This situation was also observed in the mouth of the Mafragh estuary [43]. On the other hand, the annual haline variations recorded during the period considered (2011-2014) follow the hydrological regime with sometimes drier seasons and saltier waters, and wet seasons accompanied by strong water flows and more diluted waters.

The relatively more saline waters of Station I (0,47 g/l) compared to those of Station II (0,32 g/l) could be explained by the saline and gypsiferous geological nature of the upstream region. The temporal analyzes of the period considered (2011-2014) follow relatively well the hydrology with drier seasons that generate waters on average more salty, and seasons with higher flows and limited concentrations of salts.

The variations in SM load gradually decrease from the first to the third station. Indeed, the SM content is related to the nature of the land traversed and the composition of the discharged spills.

The high SM values recorded in stations I and II can be explained by the clayey nature of the terrain and the deformation of the wadis bed that can be attributed to agricultural activity and to an intense erosion of the watershed probably the result of a brutal hydrological manifestation (floods and low flows). The most turbid waters are located upstream of the Seybouse catchment, especially in Station II, where there are numerous sand and aggregate extraction points. Although the calcium and magnesium contents can contribute to the hardness of the water (TH), the main natural sources are sedimentary rocks (loaded with calcium and magnesium) as well as soil infiltration and runoff from areas where the topsoil is thick and the rocks are calcareous with appreciable amounts of minerals Such as calcium carbonate ( $\text{CaCO}_3$ ).

Considering the high hydrometric values ( $> 54^\circ \text{f}$ ) in the three stations, we can assume that the water masses of the tributaries and the main stream of the Seybouse are considered to be very hard. Beginning in the 1970, Verneaux classified the gradients of benthic and fish fauna from upstream to downstream, based on the distance at the source, the maximum water temperature, the wetted section and the hardness of the water.

Schematically, ammonia nitrogen dissolved in water is present as ammonia ( $\text{NH}_3$ ) if the pH is greater than 8 or ammonium ( $\text{NH}_4^+$ ) if the pH is less than 8. The pH values recorded in this Study displace this dynamic equilibrium in favor of ammonia and the masses of water would have an average quality in a well-defined upstream downstream gradient. This parameter is considered lethal for ichthyofauna when pH values are less than 5 or greater than 9 [45].

The solubility of oxygen in water is closely related to temperature, atmospheric partial pressure and salinity. On the other hand, pressures from anthropogenic releases, particularly from the treatment plant adjacent to station III and the agglomeration of the city of El-Hadjar, are noticeably felt in the estuary with Dissolved oxygen which are dangerously close to total anoxia when the conditions are suitable downstream of the wadi (SIII = 4,80 mg/l). All the mean nutrient values ( $\text{NO}_3^-$ ,  $\text{NO}_2^-$ ,  $\text{NH}_4^-$ ,  $\text{PO}_4^{3-}$ ) generally indicate a mediocre to poor water mass threshold for the element under consideration. The presence of these nutrients in water at concentrations often exceeding the norms is mainly due to the boundary human activities, in particular agriculture and livestock. The situation is more worrying in the vicinity of station III which is subject to a risk of pollution by these nutrients, in particular phosphorus whose concentrations exceeds the permissible standard very much.

Perturbations of the phosphorus cycle are mainly caused by fertilizers, human and animal metabolism, and household laundry. In excessive amounts in aquatic environments, they can cause eutrophication or even dystrophication. This situation prevails in several cities in Africa where regulation is often not respected or absent [46].

The excessive presence of chlorides in the study area, particularly in the downstream station, means that the water is of poor quality. This situation indicates the contribution of anthropogenic input probably of urban as well as industrial origin. On the other hand, the sulphates are more concentrated in station III than in stations I and II, which means a dysfunction probably localized and spawned by the estuarine waters.

The analysis of variance of the morphometric and weight parameters applied to the population fractions of *B. callensis* sampled in the three stations revealed two distinct areas of occupancy. The first weakly anthropized area groups upstream stations (SI and SII) with respective barrier capture rates of 39,95% and 37,25%. This area appears to be colonized by fish of lengths and eviscerated weights average total respective of 412.6 mm (725,3 g) and 319,2 mm (334,2 g). The second area of Sidi Salem, with a catch rate of 22,8%, is occupied only by a fraction of smaller barbs ( $L_{t_{\text{way}}} = 183,8 \text{ mm}$ ,  $W_{e_{\text{wey}}} = 64 \text{ g}$ ).

Also, the position of dissolved oxygen on the positive part of the CP1 could be justified by the fact that the waters of the Seybouse basin are more oxygenated in the upstream part, far from the impact of urban effluents, which are very important downstream of the basin. At the same time, the SM content defines an axis of pollution by the natural organic and mineral particles carried by the leaching of the soils during the period of flood. It is a function of the nature of the land traversed.

On the one hand, the low mineralized waters rich in DO and SM contribute to improving the quality of the water masses and thus favor the presence of a demanding fish population. This type of region, that of the heads of basin of high and middle mountains (SI and SII), could constitute areas of refuge to this species.

On the other hand, the Sidi Salem area in the downstream part of the main stream of the maritime Seybouse is considered the most physico-chemically affected with likely impacts on the nature of the habitats frequented, the demographic structure and the dynamics of the *B. callensis* population.

Considering the salinity parameter, for example, the salinization of the downstream station (1,26 g/l) could affect the physiological metabolism of the fish, in particular their osmoregulatory function; In fact, it seems that younger individuals are more tolerant of salinity than larger individuals preferring freshwater upstream of Seybouse (<0.5 g / l).

The considerable quantities of organic matter, nutrients and minerals of domestic and industrial origin discharged upstream of station III are all factors which would favor the emergence of dystrophic seizures, resulting in the scarcity of *B. callensis* adults at the mouth of Sidi Salem to the benefit of the installation of individuals of smaller sizes and more tolerant to environmental stress.

The heterogeneity of the spatial distribution of size classes of *B. callensis* along the hydrographic network is not related solely to anthropogenic factor and adverse environmental conditions, but also to the ecobiological characteristics of the species. In fact, this species shelters in the deep water holes in winter and it is only in the spring that they are distributed [47].

However, all the results obtained do not make it possible to compare them with other bordering populations of *B. callensis*, despite its endemism in the northern part of the African continent [34, 10]. The majority of studies deal with the behavior and reaction of freshwater fishes to the influence of physico-chemical stress in a controlled environment [48-51].

The specific typology highlighted on these media, joins in its broad features the longitudinal zonation from upstream to downstream realized on the systems of running water as the one evoked [52, 53]. The work of Verneaux [54] places at the heart of the analysis the problem of the succession of bream areas (slow and hot streams), shadows and barbels (pre-mountainous regions to large rivers of plain) to trout areas marking mountain environments and cold rivers. Such a gradient has been highlighted on the Portuguese deductions [55] with nevertheless different species. These species, considered as tracers of a degradation of physico-chemical or sedimentary conditions, regress to no longer subsist downstream of the basin, as has been demonstrated in the mink *Phoxinus phoxinus* [27].

## Conclusion

In general, this study has enabled us to identify two zones which are distinct in their degree of contamination and which would significantly affect the spatial distribution in size classes of *B. callensis* along the hydrographic network. The upstream, weakly anthropized zone would host large individuals and a downstream zone on the main stream, highly mineralized and loaded with organic pollutants, consisting of very few small individuals. The high and medium Seybouse could also serve as a reservoir for fry and juveniles for downstream watersheds in difficulty (due to anthropogenic activities) and have lost their spawning stock. The knowledge gained on this species can be considered fundamental for understanding the natural mechanisms involved such as the vulnerability of this species to anthropogenic pressure. The variations observed in response to environmental stresses due to multitudes existing interrelationships between abiotic and biotic factors underline the importance of introducing this indigenous biological material into future impacts studies in continental aquatic environments.

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