# LIFE19 NAT/IT/000264

*Seagrass transplantation for transitional Ecosystem Recovery*

*(LIFE- TRANSFER)*

## Action D2: Monitoring of C2 action

## Final monitoring report

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# EXECUTIVE SUMMARY

The D.2.1 Sub-action concerns the monitoring of the growth of transplanted angiosperms. The D.2.2 Sub-action concerns the analysis of physico-chemical parameters in water samples, sediments and particulate matter, and the collection of macrophytes, benthic macroinvertebrates and fish fauna (biological elements) for the application of ecological quality indexes at the stations monitored at Comacchio (Fattibello) and Goro (Seganda, Bassunsin) lagoons.

## Rooting and Growth of Transplanted Angiosperms

Concerning the rooting and growth performance of transplanted angiosperms, no statistically significant establishment was recorded during the first year of transplantation activities (2022–2023). This outcome was primarily attributed to suboptimal environmental conditions, including elevated water turbidity in the Fattibello site and recurrent episodes of excessive algal proliferation in the Goro lagoon. During 2024, transplantation efforts were expanded to include additional lagoon sub-areas identified through preliminary assessments as having more favorable physicochemical and hydrodynamic conditions for plant establishment. In the Goro area, the Seganda station was abandoned due to poor performance, and transplantation efforts were subsequently focused on the Bassunsin station, which displayed more stable environmental conditions. Similarly, in Fattibello, the initial transplant site was discontinued, as no establishment was detected, likely due to the settlement of a large colony of flamingos that disturbed the substrate and vegetation. A new transplant site was therefore selected on the opposite side of the lagoon, where more suitable conditions for rooting and growth are expected. In the Goro area, transplantation activities involving *Ruppia cirrhosa* were further intensified, while new trials with *Nanozostera noltei*, *Zostera marina*, and *Cymodocea nodosa* were implemented in microhabitats deemed most suitable for each species' ecological requirements.

Monitoring activities carried out at the Fattibello site revealed that transplantation outcomes were not particularly satisfactory. By the end of the project, only a few isolated tufts of aquatic angiosperms were observed, indicating limited survival and establishment success of the transplanted angiosperms. These results suggest that local environmental conditions, such as sediment instability, elevated turbidity, and biotic disturbances, may have negatively affected plant rooting and persistence. Conversely, the situation at the Goro–Bassunsin site appeared slightly more favorable. Monitoring data indicated signs of *N. noltei* establishment, with sparse but distinct patches of angiosperm meadows detected, particularly in the more sheltered and protected areas of the lagoon. These findings suggest that, under suitable hydrodynamic and light conditions, the potential for successful aquatic angiosperm restoration in this area remains promising.

*Interpretation and Outlook*

The results obtained throughout the transplantation and monitoring activities highlight the strong influence of local environmental conditions on the establishment success of transplanted aquatic angiosperm species. The limited rooting and growth observed at Fattibello confirm that high turbidity levels, sediment instability, and biotic pressures—such as the disturbance caused by the presence of large bird colonies—represent key constraints to restoration success in this lagoon. These findings emphasize the need for a more detailed preliminary assessment of environmental suitability prior to transplantation, particularly regarding hydrodynamics, substrate composition, and light availability as showed by the paper of (Sfriso et al., 2023) which analyzed the relation between aquatic angiosperm presence/absence and water and sediment parameters in more than one hundred stations of the Venice Lagoon and in these stations of the Po Delta. In contrast, the partial establishment of *N. noltei* observed in autumn 2025 at the Goro–Bassunsin site demonstrates that aquatic angiosperm restoration is feasible under more favorable conditions. The positive response recorded in the most sheltered areas suggests that site selection and microhabitat characterization are critical factors determining transplant success. Future restoration actions should therefore focus on refining transplantation protocols, including the optimization of plant density, seasonality, and substrate stabilization techniques, as well as on implementing adaptive management strategies informed by continuous environmental monitoring. The experience gained during this project provides a valuable baseline for improving the design and long-term effectiveness of aquatic angiosperm restoration interventions within transitional lagoon ecosystems.

**Macrophytes and MaQI index**

Macrophytes are the organisms selected by the Water Framework Directive (Directive 2000/60/EC) that respond most quickly to a change in ecological status, as they can change their typology within a few months. Therefore, the response is very rapid and relies on both the presence of species of high ecological value (sensitive taxa) and the typology and cover of aquatic angiosperms.

To take into account winter and summer species, the assessment of ecological status is based on two surveys of aquatic vegetation: one in spring (generally May) and one in autumn (generally November), and considers all macroalgal species and the maximum cover of macroalgae and aquatic angiosperms detected in the two surveys.

These samplings of all the macroalgal variables carried out every year from 2022 both in Goro and Fattibello did not show the presence of algal species of high ecological value but since autumn 2025 at Goro the rooting of *Nanozostera noltei* that formed small patches was observed increasing the ecological status from Poor to Moderate. Instead, at Fattibello the presence of scattered small tufts of *Ruppia cirrhosa* ,that have not yet developed, were recorded. Thereore, the ecological status remained Poor but giving hope for their expansion and consequent environmental improvement.

**Macrobenthos and M-AMBI and BITS index**

Concerning the benthic fauna, 37 macrobenthic taxa were found in the Sacca di Goro, and 33 in Valle Fattibello. The ecological quality, as assessed through macrobenthic community indices during the *ex-ante* monitoring phase, was found to be higher at the control sites compared to the transplant sites, both in Goro and Fattibello. This pattern indicated initially better ecological conditions in the non-intervention areas. However, throughout the project, monitoring data revealed a slight but consistent improvement in ecological quality at the transplant sites relative to the controls. This trend suggests a progressive enhancement of benthic community structure and diversity in correspondence with the restoration activities. Although the variation remains moderate, the observed increase in macrobenthic index values at the transplant sites may reflect an initial recovery of habitat conditions and an early ecological response to aquatic angiosperm establishment. The majority of the macrobenthic taxa present at all the sites belongs to the indifferent (EG-II) and tolerant (EG-III) group at Goro, and tolerant and opportunistic (EG-IV) group at Fattibello.

**Fish Fauna and HBFI Index**

The application of the HBFI (Habitat-Based Fish Index) produced contrasting results between the two lagoons, which are considered to be primarily related to their different geomorphological characteristics rather than to the transplantation activities. In the Goro lagoon—an open-type system, with the transplant site located close to the sea inlet—the environmental quality consistently ranged from *Good* to *High* status, both at the transplant and control sites. Conversely, in the Fattibello lagoon—a choked system with limited water exchange—the environmental quality was consistently assessed as *Moderate* or even *Poor*. These outcomes confirm that the intrinsic hydromorphological features of each lagoon play a major role in determining fish assemblage structure and, consequently, the HBFI-based ecological status, often overriding the effects of localized restoration actions. These findings highlight the importance of considering lagoon typology and hydrodynamic connectivity when evaluating restoration effectiveness and selecting future intervention sites, ensuring that ecological improvements are assessed within an appropriate environmental context.

**Conclusions and Recommendations**

The transplantation activities produced mixed results across the two lagoons. At Fattibello, environmental constraints, including high turbidity and disturbance from a large flamingo colony, limited the survival and establishment of transplanted angiosperms. In contrast, Goro–Bassunsin showed partial success, with *N. noltei* establishing in sheltered areas, highlighting the importance of careful site selection. Macrobenthic indices indicated that transplant sites initially had lower ecological quality than control sites, but slight improvements were observed over the course of the project, suggesting a positive response of benthic communities to restoration efforts. HBFI-based fish assessments showed that lagoon morphology strongly influenced ecological status, with the open Goro lagoon maintaining good to high environmental quality, while the choked Fattibello lagoon remained moderate to poor. The key recommendations can be summarized as: (i) *focus on suitable sites*, i.e. prioritize areas with stable hydrodynamics, good water quality, and appropriate substrates to maximize transplant success; (ii) *careful species selection,* i.e. choose the most suitable species for each site; at Goro, four species were tested, but only one showed signs of successful establishment; (iii) *adaptive management*, i.e. continue monitoring and adjust restoration strategies in response to environmental changes; (iv) *stakeholder engagement*, i.e. continue to collaborate with local fisherman communities and organizations to ensure long-term sustainability. Implementing these recommendations will support more effective angiosperm restoration, enhance benthic and fish community health, and contribute to the long-term conservation of Goro and Fattibello lagoon ecosystems..

# SubAction D.2.1 Monitoring angiosperm growth

Aquatic angiosperms at the beginning of the project were absent in both the lagoons. The only species present in the past was *Ruppia cirrhosa*, but it disappeared in the last three decades of the last century with the increasing of eutrophication (Sfriso et al., 2016; Munari et al., 2023).

This project aims to reintroduce this species and/or others aquatic angiosperms present in similar environments to reconstruct, where possible at least in part, the ancient prairies.

**Table 1** shows the number of transplants foreseen in each lagoon in the Proposal for a total of 648 sods equivalent approx. to 6480 rhizomes.

***Table 1***. *Number of planned transplants in each lagoon.*

| Year | 2021 | | 2022 | | 2023 | | 2024 | | 2025 | | Total |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Season | --- | Autumn | Spring | Autumn | Spring | Autumn | Spring | Autumn | Spring | Autumn | |
| Stations | --- | 8 | 8 | 8 | 8 | 8 | 8 | 8 | 8 | 8 | 72 |
| N° of sods | --- | 72 | 72 | 72 | 72 | 72 | 72 | 72 | 72 | 72 | 648 |
| Equivalent rhizomes | --- | 720 | 720 | 720 | 720 | 720 | 720 | 720 | 720 | 720 | 6480 |

**Table 2** shows the transplants carried out at Goro and Fattibello, indicating the number of sods transplanted per year and the corresponding number of equivalent rhizomes, while **Figures 1** and **2** show transplant activities at both lagoons. Overall, the total number of sods and rhizome-equivalents transplanted exceeded the original targets.

***Table 2.*** *Number of effective transplants*

| N° of transplanted sods and rhizomes | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Year | Species | Sods (diameter 15 cm) | Equivalent rhizomes | Donor | Rhizomes | Equivalent Sods | TOTAL Sods | TOTAL Rhizomes |
| Goro | 2021 | *R. cirrhosa* | 207 | 3519 | Valle Bertuzzi | - | - | | |
| | 2022 | *R. cirrhosa* | 212 | 3604 | Valle Bertuzzi | - | - | | |
| | | *C. nodosa* (UniVE) | - | - | Venice Lagoon | 100 | 6.7 | | |
| | | *Z. marina* (UniVE) | - | - | Venice Lagoon | 300 | 30.0 | | |
| | | *N. noltei* (UniVE) | - | - | Venice Lagoon | 600 | 40.0 | | |
| | 2023 | *Z. marina* | - | - | Venice Lagoon | 150 | 15.0 | | |
| | | *N. noltei* | - | - | Venice Lagoon | 350 | 23.3 | | |
| | | *R. cirrhosa* | 72 | 1224 | Valle Bertuzzi | - | - | | |
| | | *R. cirrhosa* | 72 | 1224 | Valle Bertuzzi | - | - | | |
| | | *R. cirrhosa* | 10 | 170 | Valle Bertuzzi | - | - | | |
| | 2024 | *R. cirrhosa* | 50 | 850 | Valle Bertuzzi | - | - | | |
| | | *N. noltei* | 400 | 6000 | Venice Lagoon | - | - | | |
| | 2025 | *N. noltei* | 750 | 11250 | Venice Lagoon | - | - | | |
| SubTotal | | | 1773 | 27841 | | 1500 | 115 | 1888 | 29341 |
| Fattibello | 2021 | *R. cirrhosa* | 80 | 1360 | Valle Campo | - | - | | |
| | 2022 | *R. cirrhosa* | 140 | 2380 | Valle Bertuzzi | - | - | | |
| | 2023 | *R. cirrhosa* | 140 | 2380 | Valle Bertuzzi | - | - | | |
| | 2024 | *R. cirrhosa* | 80 | 1360 | Valle Bertuzzi | - | - | | |
| | 2025 | *R. cirrhosa* | 150 | 2550 | Valle Bertuzzi | - | - | | |
| SubTotal | | | 590 | 10030 | | - | - | 590 | 10030 |

***Figure 1.*** *Transplant activities at Goro*

***Figure 2.*** *Transplant activities at Fattibello*

## Angiosperm rooting and growth monitoring

The monitoring of the establishment and growth of the transplanted angiosperms was carried out twice a year (usually in spring and in late summer–early autumn) from 2022 until September 2025. As previously mentioned, despite the continuous transplanting efforts carried out by fishermen and DAIS-UNIVE staff, as of September 2025 only small, scattered tufts of angiosperm meadow are present at the Goro-Bassunsin site, and only very sparse tufts at the Fattibello site, indicating very limited establishment success.

It is well known that *Ruppia cirrhosa* can experience periods of limited vegetative growth, while remaining viable through its rhizomes. In the Fattibello site, it is therefore expected that a considerable number of living rhizomes are still present, providing the potential for future expansion of the above-ground vegetation under favorable conditions.

In the Goro lagoon, however, *Ruppia* did not establish successfully. The patches observed there, totaling several tens of meters in extent, are composed of *Nanozostera noltei*. Similarly, it is anticipated that these patches may expand orthotopically over time, and potentially release seeds that could facilitate further colonization of the substrate, contributing to the gradual recovery of angiosperm meadows in suitable microhabitats.

Continued monitoring of both vegetative growth and rhizome persistence will be essential to evaluate long-term restoration success. Understanding seasonal dynamics and reproductive potential will help refine transplantation strategies and support adaptive management, ultimately enhancing the resilience and expansion of angiosperm communities within the lagoons.

Following **Figure 3** shows *N. noltei* patches at Goro-Bassunsin in September 2025.

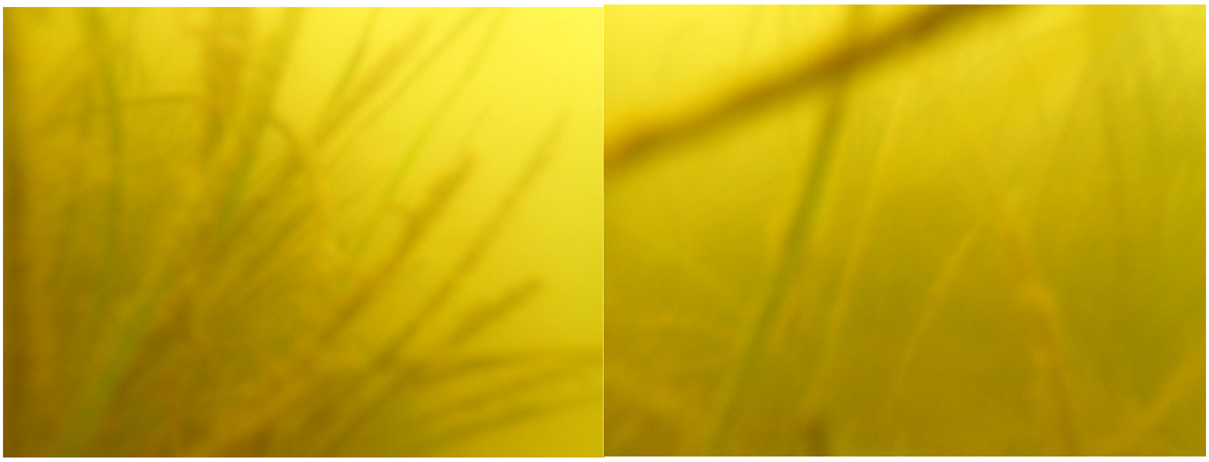


***Figure 3****. Nanozostera noltei patches in September 2025 (with very turbid water)***.** *The diameter was 30-40 cm.*

The following **Figures 4** and **5** summarize the outcome of the transplants in each lagoon.


***Figure 4***. *Goro lagoon*


***Figure 5***. *Fattibello lagoon*

# Sub-action D2.2 Monitoring the environment and the ecological quality status

## SECTION 1 – Environmental parameters: water and sediments

***Methods***

*Field sampling*

Once the ecologically suitable sites had been selected for the transplantation of aquatic angiosperms, the sampling of the environmental matrices of water and sediments started. In each station some environmental parameters were monitored by means of portable instruments in accordance with what was reported in the sampling forms.

In field the following parameters and macrophyte variables were recorded:

- Date and time of the surveys;
- Air and water temperature;
- Water depth and transparency by Secchi disk;
- Dissolved oxygen;
- pH and Eh in the water column by dedicated probe for water measures;
- pH and Eh in the surface sediments by dedicated probe for sediment measures;
- Water samples for salinity determination;
- Water samples for nutrient determination;
- Sediment samples for physico-chemical and nutrients determination;
- Macroalgal coverage, macroalgal biomass, taxa dominance, samples to determine the complete macroalgal check list.

Water samples (250-500 ml) were manually collected at a depth of approx. 20-30 cm and immediately filtered throughout glass fiber filters GF/F (porosity 0.7µm). Filters were retained in filter-holder boxes until the determination of Chlorophyll-*a* (Chl-*a*) and Phaeopigments (Pheo-*a*). Water samples of 250 ml were retained in polyethylene bottles for the determination of nutrients and transported in laboratory by a fridge bag. Both filters and water samples were refrigerated until the determination.

Surface sediment samples (5 cm top layer) were collected by a manual Plexiglas corer (Ø=10cm). They were carefully mixed together and pH and Eh were immediately measured on the total homogenized sample. Then two subsamples were retained, one for the analysis of nutrients and the other for the determination of the sediment characteristics. Sediment samples were transported by fridge bag to the laboratory where they were frozen and lyophilized for the determination of fines (fraction <63µm), density, moisture, porosity and the total, inorganic, organic phosphorus, total, inorganic, organic carbon and total nitrogen.

***Measures in water column and surface sediments***

***Temperature***

The determination of the temperature in the water column at a depth of approx. 30 cm was obtained by means of a thermocouple probe (precision 0.1° C) combined with a portable pH-meter model Delta Ohm HD8705.

***Dissolved oxygen***

The determination of dissolved oxygen (OD) at about 20-30 cm depth was carried out using an Oximeter (OXI 196) from Wissenschaftlich-Technische Werkstätten GmbH (Germany). The data expressed instrumentally in mg $L^{-1}$ were then converted into saturation percentage (%OD) taking into account the temperature and salinity values. The instrument was calibrated before each series of measurements in its wet container.

***pH determination***

The determination of the pH (acidity or basicity or neutrality) in the water column was carried out using the Delta Ohm HD8705 portable pH-meter, equipped with a combined electrode (accuracy 0.01 pH units). The instrument was calibrated before each sampling campaign with a pH 7.0 solution.

***Redox potential determination***

The determination of the red-ox potential in the water and in the surface, sediment was carried out by means of a Delta Ohm HD8705 portable pH-meter equipped with a combined Ag (AgCl) electrode (precision 1 mV). The measurement in water was carried out at a depth of 20-30 cm while in the sediment it was carried out on a sample of 3 sub-samples (5 cm top layer), carefully homogenized, collected by means of a Plexiglas corer (Ø=10cm). The measurement on the homogenized sample avoids the enormous variations that occur depending on very small variations in the insertion of the electrode into the superficial sediment.

***Salinity determination***

Salinity was determined in laboratory as chlorinity by means of Oxner (1962) argentometric titration. The chlorinity values corrected with a standard solution of sea water of known chlorinity were converted into salinity by means of the relationship: Salinity = $Cl^-$ x 1.805 + 0.03.

***Suspended solids (filtered particulate matter) determination***

Samples of the water column (250-500 ml) were filtered, in duplicate, throughout glass fiber filters GF/F (0.7μm) pre-dried at 105 ° C for 1 hour and weighed for the measurement of the total suspended solids (TSS). After filtration by a Millipore Swinnex manual apparatus the samples were washed with 2-3 aliquots of distilled water (20 ml) to remove the salts. Filters were placed in filter-holder boxes and refrigerated until the moment of the determination which took place by drying in an oven at 70 ° C for one night. The coefficient of variation as a measure of reproducibility of the analysis was kept below 5%.

***Nutrients and chlorophylls in water***

The filtered seawater was analysed for nutrient concentrations following the methods described in Strickland & Parsons (1972) whereas chlorophylls and phaeopigments were determined according to the Lorenzen (1967) method.

Briefly, phytoplankton concentration was determined as Chl-*a* and Pheo-*a* by extraction with acetone 90% in ultrasonic bath for 30 min. Reactive phosphorus (RP) was measured as in Murphy & Riley (1962) and reactive silicate (Si) was quantified using the reaction of Mullin & Riley (1965). Ammonium was measured with the phenol-hypochlorite reaction of Riley (1953) modified by Solarzano (1969). The simultaneous determination of nitrite and nitrate concentrations was obtained using the cadmium reduction method as in Wood et al. (1967). The results were expressed as μg $L^{-1}$ for chlorophylls and as μM for nutrients.

***Sediment grain-size determination***

Sediments were sieved with a 1 mm mesh sieve to remove the coarser part mainly represented by shell residues. Then the fine fraction (Fines) and the sands were separated with a 63 μm mesh sieve.

### *Sediment density*

The sediment density was determined on wet and dry basis. In particular the values on dry basis (g/cm$^3$) allow to calculate the concentration of nutrients or pollutants per volume unit.

The density determination was performed in duplicate by using porcelain crucibles of known volume comparing the sediment weight before and after drying at 110 °C for one night.

### *Sediment Moisture and Porosity*

The same porcelain crucibles of known volume were also used to determine the sediment moisture (ml of water/ weight of wet sediment) and porosity (ml of water/ volume of wet). All data are reported as a percentage.

### *Elemental Analysis of C, N and P*

In the laboratory, sediments were freeze-dried and pulverized using a sediment mill (Fritsch Pulverisette, Germany). The concentration of total nitrogen (Ntot) and total carbon (Ctot) were measured in duplicate by a CHNS Analyzer (Vario-MICRO, Elementar CHNS by Elementar Italia S.r.l.) after an accurate sample powdering of ca. 0.3 g of sample. The standard used for nitrogen determination was the "low level N- and S-contents" with N = 0.74%, art. no. 05 000 959 (Elementar Italia S.r.l.) and the standard used for carbon determination was "C2*", with C = 2.00%,* art. no. S05 005 343 (Elementar Italia S.r.l.). Organic Carbon (Corg) was measured as carbon loss on ignition after combustion at 430 °C for 2h taking into account the weight loss of the sample following the combustion.

Total phosphorus (Ptot) was determined following Aspila et al. (1976) after sample combustion in the muffle at 550 °C for at least 2 h of 0.3–0.4 g of sample. Subsequently, the residue thus obtained was suspended in 50 mL of 1 N HCl and sonicated for ca. 30 min. After allowing the sample to settle for at least 1 h, 0.5 mL of the supernatant were taken with a graduated gaschromatographic syringe and brought to exactly 10 mL using volumetric flasks for a final dilution of 1 L, with the result expressed directly in µM. At this point, the phosphorus concentration was determined spectrophotometrically by the molybdenum blue method adding the mixed reagent and reading the absorbance at 885 nm after ca. 10–15 min according to Murphy et Riley (1962) and Strickland et Parsons (1972). Inorganic Phosphorus (Pinorg) was obtained with the same procedure used for Ptot but without combustion at 550 °C. Organic phosphorus (Porg) was determined by difference.

All samples were analysed in duplicate, and the analyses were replicated on two different days to obtain an accuracy > 95. Otherwise, the analyses were repeated until the coefficient of variation (standard deviation/mean) between two replicates was <5%. Carbon and Nitrogen contents were expressed as mg $g^{-1}$ and Phosphorus as μg $g^{-1}$.

**Results**

- ***<u>EX-ANTE MONITORING</u>***

**Water Column**

*Samples were recorded just once on 06/07/2021 at Goro and on 27/05/2021 at Fattibello*

***Water parameters***

Some physico-chemical parameters of the water column are shown in **Table 3.**

***Table 3.*** *Water column parameters.*

| Water column | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Stations | Physico-chemical parameters | | | | | | | | | |
| | Temperature | | pH | Eh | Salinity | DO | Depth | Transparency | | TSS |
| | air | water | | mV | psu | % | cm | cm | % | |
| Goro | 23.2 | 19.2 | 8.30 | 198 | 6.7 | 82.4 | 30 | 30 | 100 | 52 |
| Fattibello | 27.1 | 19.3 | 8.20 | 308 | 19.2 | 96.9 | 30 | 20 | 67 | 81 |

Temperature showed the typical values of the considered season. The pH values ranged between 8.20 and 8.30 units whereas the redox potential ranged between 198 mV and 308 mV. Salinity at Goro was only 6.7 psu whereas at Fattibello it was 19.2 psu.

***Table 4.*** *Nutrient concentrations in the water column.*

| Nutrients in the water column | | | | | | |
|---|---|---|---|---|---|---|
| | RP | $NH_4^+$ | $NO_2^-$ | $NO_3^-$ | DIN | SI |
| Goro | 0.37 | 11.2 | 2.64 | 25.2 | 39.0 | 24.8 |
| Fattibello | 0.41 | 2.07 | 1.35 | 2.79 | 6.21 | 34.5 |

The concentrations of Reactive Phosphorus (RP) where relatively low at both stations with the highest value at Fattibello (0.41 μM) and a concentration slightly lower at Goro.(0.37 μM) (**Table 4**). The concentration of the Dissolved Inorganic Nitrogen (DIN = sum of ammonium, nitrite, nitrate) was significantly higher at Fattibello (39.0 μM) than at Goro (6.21 μM) due to the highest concentrations of Nitrates (25.2 µM). Conversely silicates (Si) where higher at Fattibello (34.5 µM) than at Goro (24.8 µM).

### *Surface sediments*

The results of some physico-chemical properties of sediment samples are reported in **Table 5.**

**Table 5**. Physico-chemical characteristics in the 5 cm top layer

| Sediment properties | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Stations | Shells >1mm | Sand >63µm | Fines <63 µm | Density | | Moisture | Porosity | 5 cm top layer | |
| | | | | dry | wet | | | | |
| | | % | | g cm$^{-3}$ | | % | | pH | Eh (mV) |
| Goro | 1.27 | 4.05 | 94.7 | 0.57 | 1.35 | 57.6 | 77.9 | 7.14 | -126 |
| Fattibello | 0.89 | 43.1 | 56.0 | 0.85 | 1.52 | 43.8 | 66.4 | 7.32 | -187 |

The sediment 5 cm top layer showed physico-chemical characteristics quite different. The amount of dry sediment per volume unit (dry density) ranged between 0.57 g cm$^{-3}$ at Goro and 0.85 g cm$^{-3}$ at Fattibello. The sediment grain-size <63 µm (Fines) was reversed with the highest concentration at Goro (94.7%) compared to Fattibello (56.0%). As consequence Moisture and Porosity were higher at Goro: 57.6 and 77.9%, respectively, than at Fattibello (43.6 and 66.4%). Sediment pH was in the range 7.14-7.32 units whereas Eh was negative with values lower at Fattibello (-187 mV).

**Table 6**. Nutrient concentrations in 5 cm surface top layer

| Nutrient in surface sediments | | | | | | | |
|---|---|---|---|---|---|---|---|
| Stations | Ptot | Pinorg | Porg | Ntot | Ctot | Cinorg | Corg |
| | | µg g$^{-1}$ | | | mg g$^{-1}$ | | |
| Goro | 787 | 605 | 182 | 2.1 | 37.2 | 16.2 | 21.0 |
| Fattibello | 688 | 628 | 60 | 1.1 | 32.2 | 24.0 | 8.2 |

As expected by considering the highest amount of Fines sediments at Goro the concentration of Total Phosphorus (Ptot = 787 µg $g^{-1}$, **Table 6**) was higher than at Fattibello (688 µg $g^{-1}$) due to the contribution of Porg (182 µg $g^{-1}$) that was approximately three times higher than at Fattibello (60 µg $g^{-1}$). Similar results were recorded for Total Carbon (Ctot) where the contribution of the organic fraction was determinant.

Higher concentrations were found at Goro also for Total Nitrogen (TN) which showed a concentration of 2.1 mg $g^{-1}$, value significantly higher than at Fattibello (1.1 mg $g^{-1}$).

***Primary producers***

The concentration of phytoplankton was determined by measuring the total Chlorophyll-*a* (Chl-*a* tot) composed by the active Chlorophyll-*a* (Chl-*a*) and the inactive pigment, namely phaeopigment (Phaeo-*a*) (**Table 7**).

**Table 7**. Chlorophyll-*a* concentrations in the two stations

| Stations | Chl-*a* | Pheo-a | Chl-*a* tot |
|---|---|---|---|
| Goro | 1.08 | 5.36 | 6.44 |
| Fattibello | 25.1 | 7.54 | 32.7 |

On the whole the concentration of total (Chl-*a* tot) was very high in both stations (**Table 8**) and at Fattibello reached even 32.7 µg $L^{-1}$ with the active pigment (Chl-a) that prevailed on the inactive one (Phaeo-a). At Goro the total Chl-*a* was very lower (6.44 µg $L^{-1}$) and, in this case, Phaeo-*a* (5.36 µg $L^{-1}$) was approximately 5 times higher than Chl-*a* (1.08 µg $L^{-1}$).

No aquatic angiosperms were present in the selected stations whereas macroalgae represented mostly by *Ulva australis* and Gracilariaceae showed a significant biomass at Fattibello (1695 g $m^{-2}$) and negligible at Goro (2.9 g $m^{-2}$) (**Table 8**).

The number of taxa recorded in the four lagoons was very low: two taxa at Goro (1 Chlorophycea, 1 Rhodophycea) and three taxa at Fattibello (1 Chlorophycea, 2 Rhodophyceae. None of the species was of high ecological value (sensitive taxa).

**Table 8**. *Ex-ante* macroalgal taxonomic list and some metrics. In green: Chlorophyceae, in red: Rhodophyceae.

| Taxonomic list *ex-ante* | | | |
|---|---|---|---|
| N° | Macroalgae | Goro | Fattibello |
| 1 | ***Ulva laetevirens*** Areschoug | x | x |
| 2 | ***Agardhiella subulata*** (C. Agardh) Kraft *et* M.J. Wynne | | x |
| 3 | ***Agarophyton vermiculophyllum*** (Ohmi) Gurgel, J.N. Norris et Fredericq | x | |
| 4 | ***Gracilariopsis longissima*** (S.G. Gmelin) Steentoft *et al.* | | x |
| | **Total taxa** | **2** | **3** |
| | **Chlorophyceae N°** | 1 | 1 |
| | **Rhodophyceae N°** | 1 | 2 |
| | **Phaeophyceae N°** | 0 | 0 |
| | **Cover %** | **10** | **70** |
| | **Biomass g/m²** | **2.9** | **1695** |
| | **Chlorophyceae%** | **85** | **99** |
| | **Rhodophyceae%** | **15** | **1** |
| | **Chlorophyceae abundance** | **25.5** | **69.3** |
| | **Rhodophyceae abundance** | **4.5** | **0.7** |
| | **MaQI** | **0.25** | **0.25** |

Since both stations lacked aquatic angiosperms and macroalgae of high ecological value, the ecological status determined by applying the Macrophyte Ecological Index (MaQI, Sfriso et al., 2014) was "Poor" with score 0.25 due to the dominance of Chlorophyceae.

**Discussion and Conclusions**

The *ex-ante* monitoring of the two lagoons allowed to characterize the ecological conditions of these basins both from the physico-chemical and biological point of view. Overall, the areas selected in two lagoons presented sediments prevalently muddy. In general, the concentration of RP in the water column was relatively low whereas DIN was high especially at Goro due to the high contribute of nitrates. Vice versa, surface sediments showed a high concentration of Ptot especially at Goro due to the contribution of Porg.

The lack of aquatic angiosperms and sensitive macroalgae, and the low macrophyte biodiversity highlighted Poor ecological conditions.

- ***TWO YEAR MONITORING ON MONTHLY BASIS***

In April 2025 we concluded the second year of monthly sampling both at Goro and Fattibello. Some images are reported in **Figure 6** and **Figure 7.**

-

***Figure 6.*** *On the left lagoon of Goro with reeds indicating the transplant points of Ruppia cirrhosa. On the right trap for the collection of the particulate matter.*

***Figure 7.*** *On the left, sampling operation on the lagoon of Fattibello. On the right, equipment and materials for sampling operations*

The mean water variations of Temperature, pH, Eh, Salinity, Oxygen Saturation, Total Suspended Solids in the water column is shown in **Figure 8**.

The mean annual water temperature at Goro station was 18.5±8.5 °C in  2022-23 and 18.4±8.0 °C in 2024-25 with a men value of 18.4±8.1 °C. At Fattibello it was slightly higher ranging from 20.4±8.6 °C in 2022-23 to 19.5±8.5 °C in 2024-25, with a mean value of 20.0±8.4 °C.  The *ex-ante* values recorded in May 2021 were within this range.

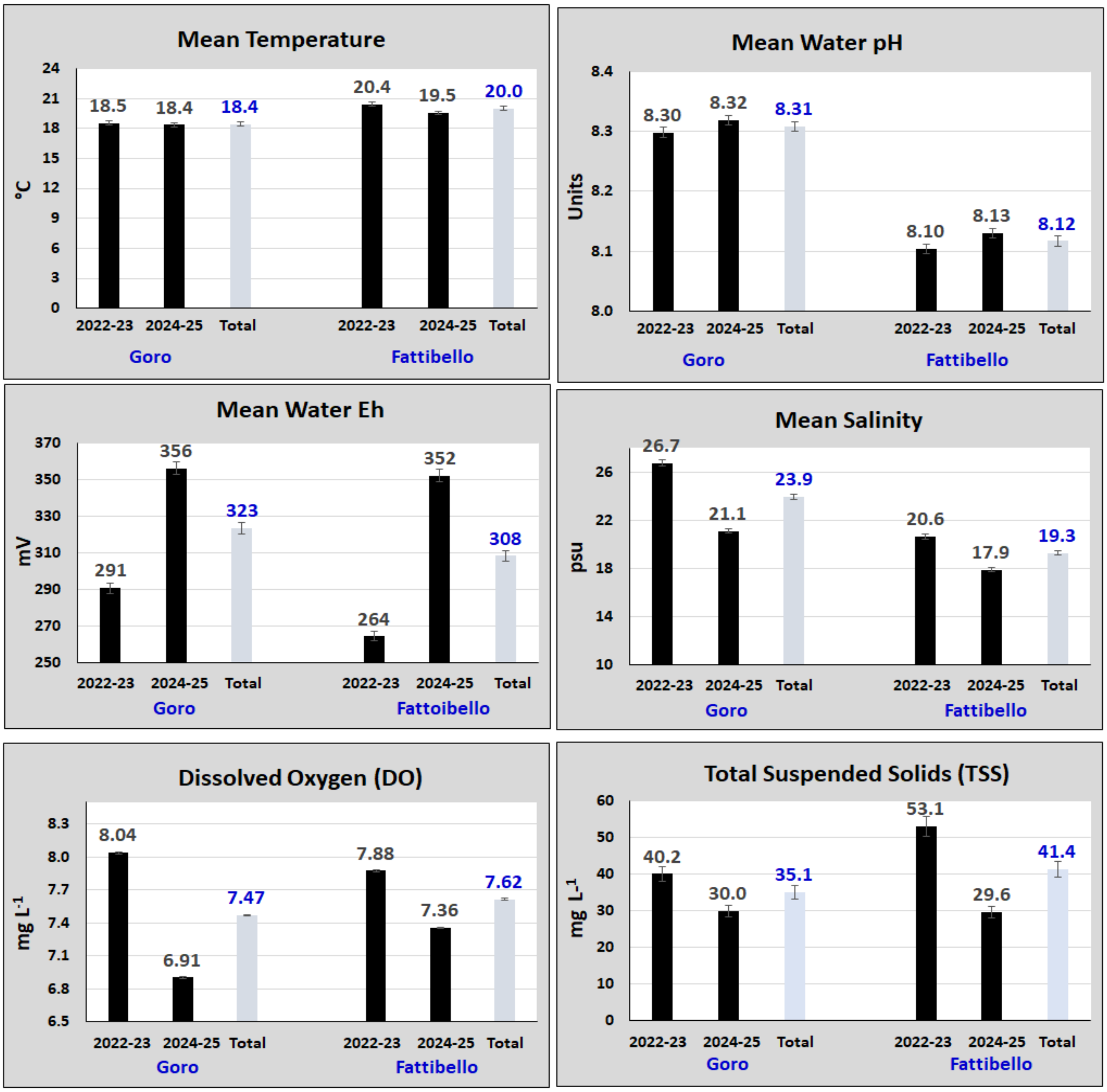



***Figure 8.*** *Variation of some parameters of the water column during the two sampling years.*

Temperature changes on monthly basis are reported in **Figure 9**. At Goro the minimum value was recorded in January 2023 (6.6 °C) and the maximum in July 2024 (29.9 °C). At Fattibello temperature were slightly higher ranging from 6.9 °C in December 2024 to 30.6 in August 2024..

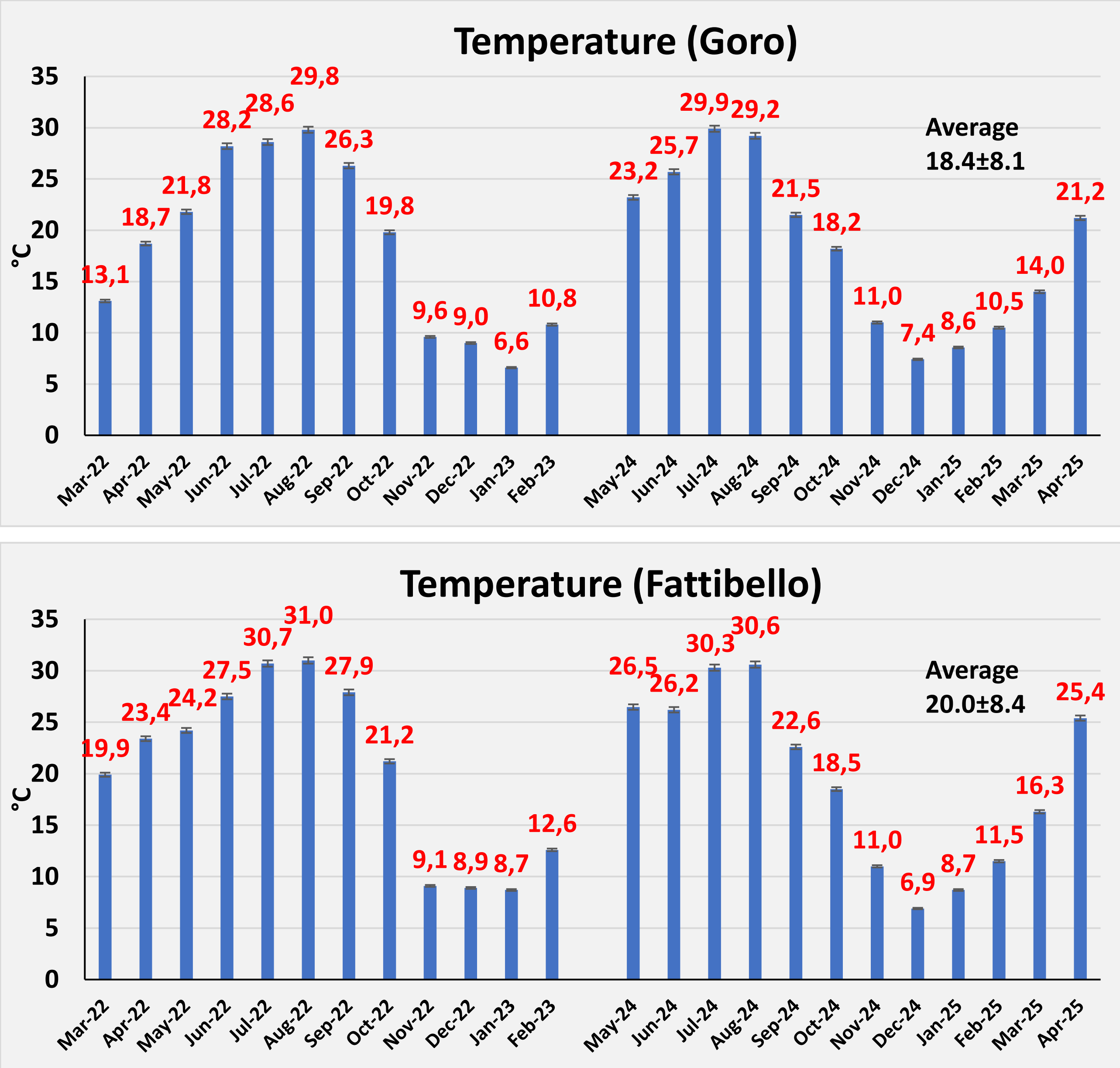



***Figure 9.*** *Variation of temperature at Goro and Fattibello in the water column during the two sampling years.*

The mean values of pH at Goro ranged from 8.30±0.37 in 2022-23, to 8.32±0.20 in 2024-25 with a mean value of 8.31 (**Figure 8**). At Fattibello pH was lower ranging from 8.10±0.20 to 8.13±0.28 with a mean value of 8.12. The *ex-ante* values were within these ranges.

Similarly, the mean redox potential (Eh) at Goro was 291±56 mV in 2022-23 and 356±27 mV in 2024-25, with a mean value of 323 mV, whereas at Fattibello the mean Eh ranged from 264±100 mV in 2022-23 to 352±19 mV in 2024-25 with a mean range of 308 mV.

Salinity mean values were higher at Goro (23.9±5.9 psu) than at Fattibello (19.3±6.8 psu) (**Figure 8**). They ranged between 9.5 psu in May 2024 and 30.8 psu in August 2022  at Goro and from 2.7 psu in October 2024 to 29.5 psu in July 2022 at Fattibello (**Figure 10**). The salinity recorded in the *ex-ante* at Goro and Fattibello were 6.7and 19.2 psu, respectively, but it was strongly affected by the Po river inputs in the different years.

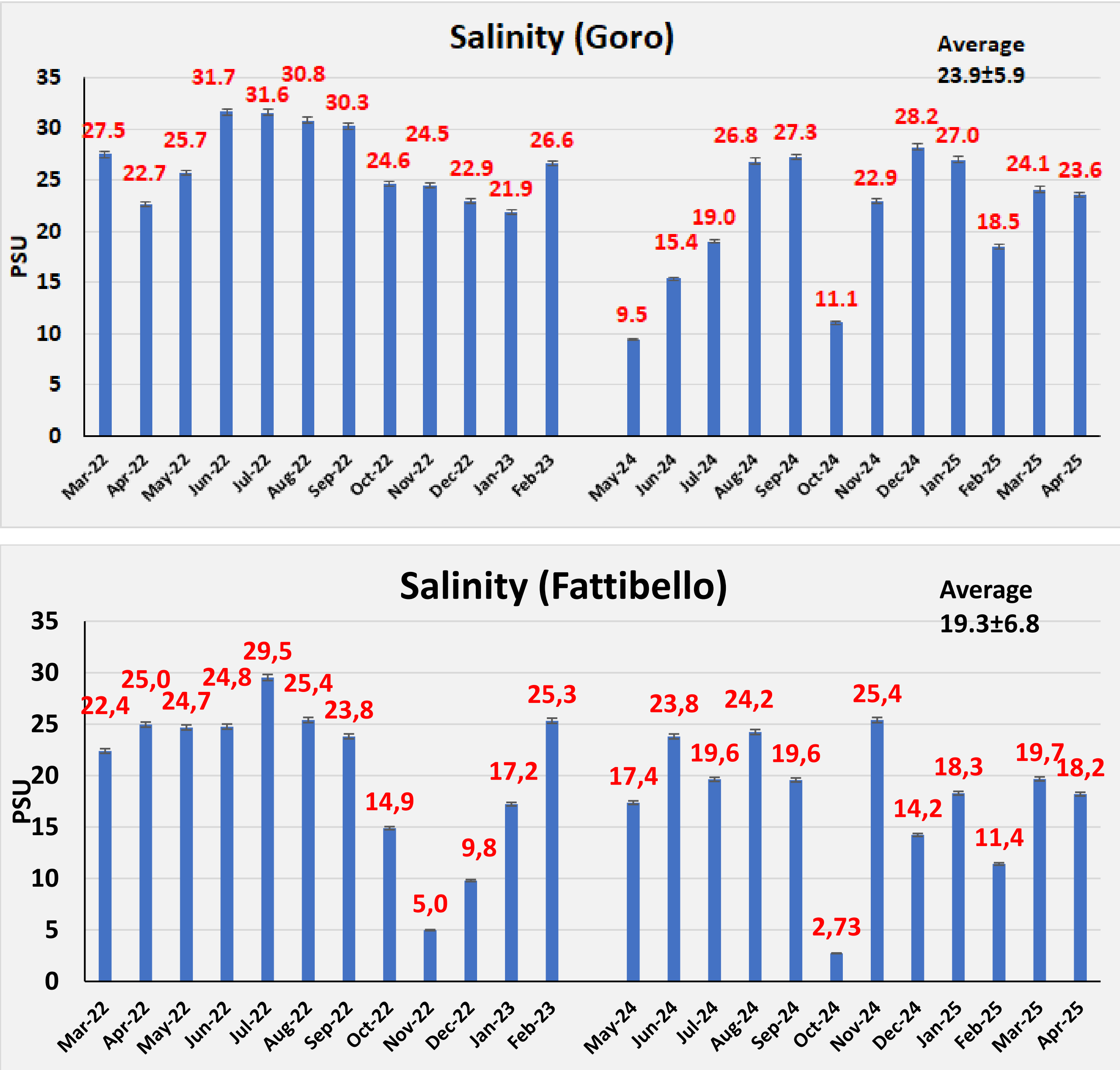
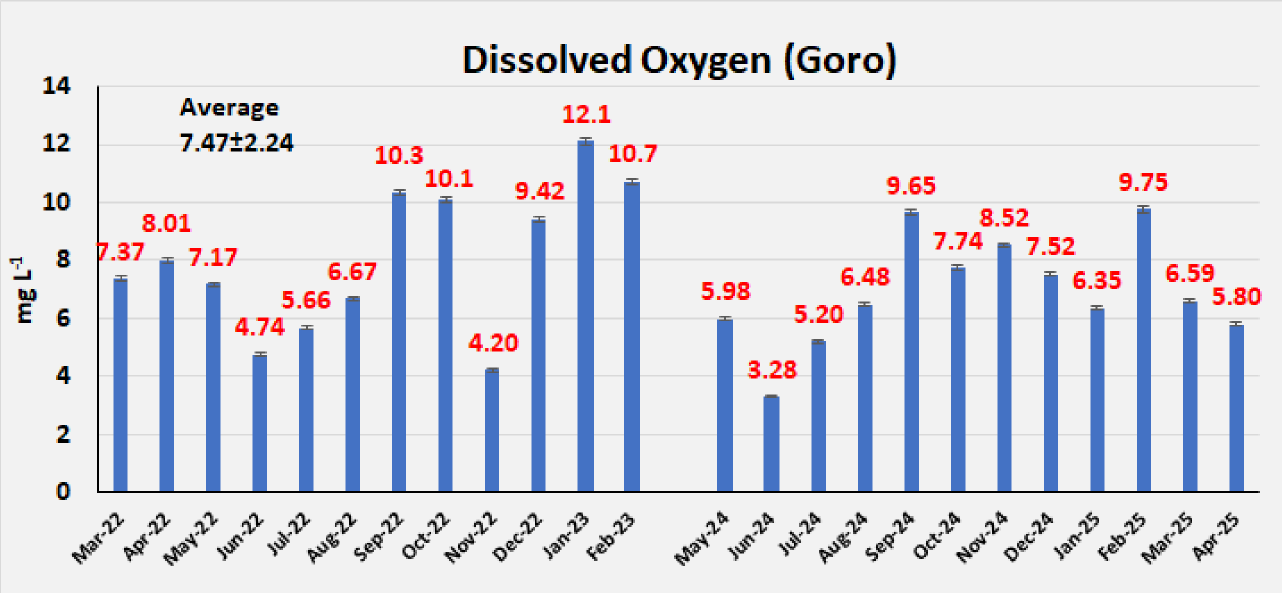



***Figure 10.*** *Variation of some environmental parameters in the Water Column in the Goro and Fattibello stations.*

The total average Dissolved Oxygen (DO) concentration recorded over the two years at Goro (7.47±2.24 mg $L^{-1}$) was slightly lower than at Fattibello (7.62±2.21 mg $L^{-1}$) (**Figure 8**). At Goro in 2022-23 the mean concentration was 8.04±251 mg $L^{-1}$ and decreased to 6.91±1.86 mg $L^{-1}$ in 2024-25. In contrast, at Fattibello the mean DO concentration in 2022-23 it was 7.88±2.54 whereas in 2024-25 it decreased to 7.36±1.91 mg $L^{-1}$. The DO monthly values ranged from 3.28 mg $L^{-1}$ in June 2024 to 12.1 mg $L^{-1}$ in January 2023 at Goro (**Figure**

**11**). Instead at Fattibello the lower DO concentration (3.22 mg L$^{-1}$) was recorded in July 2022 and the highest (11.1 mg L$^{-1}$) in February 2023. The values recorded in the *ex-ante* monitoring were different within this range.



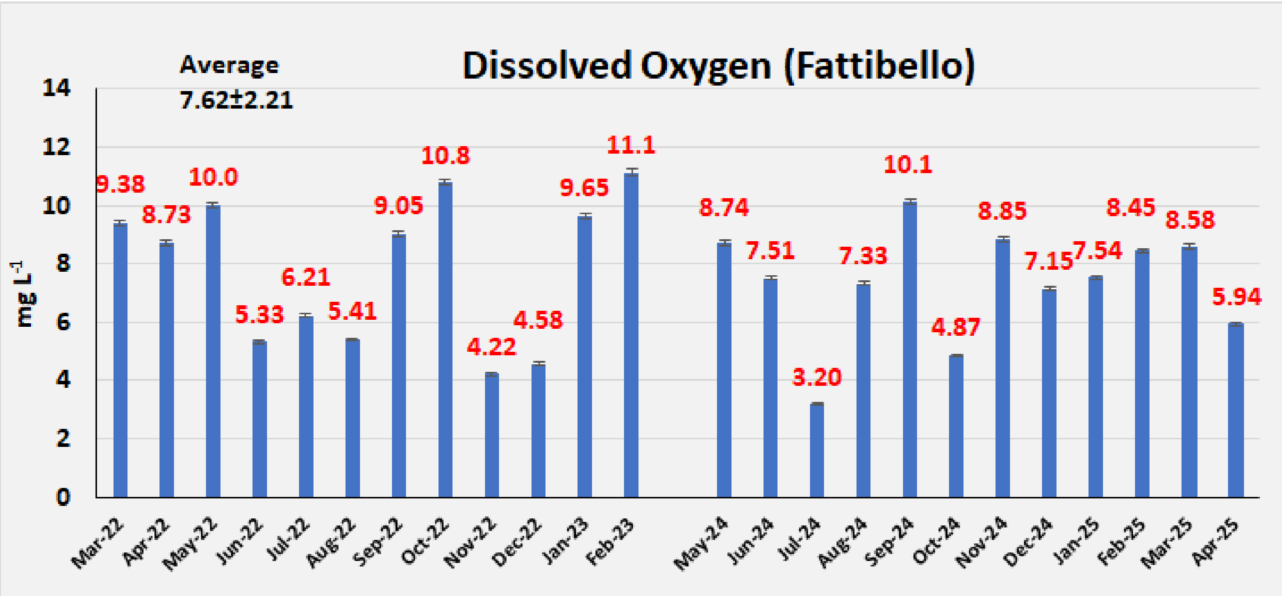
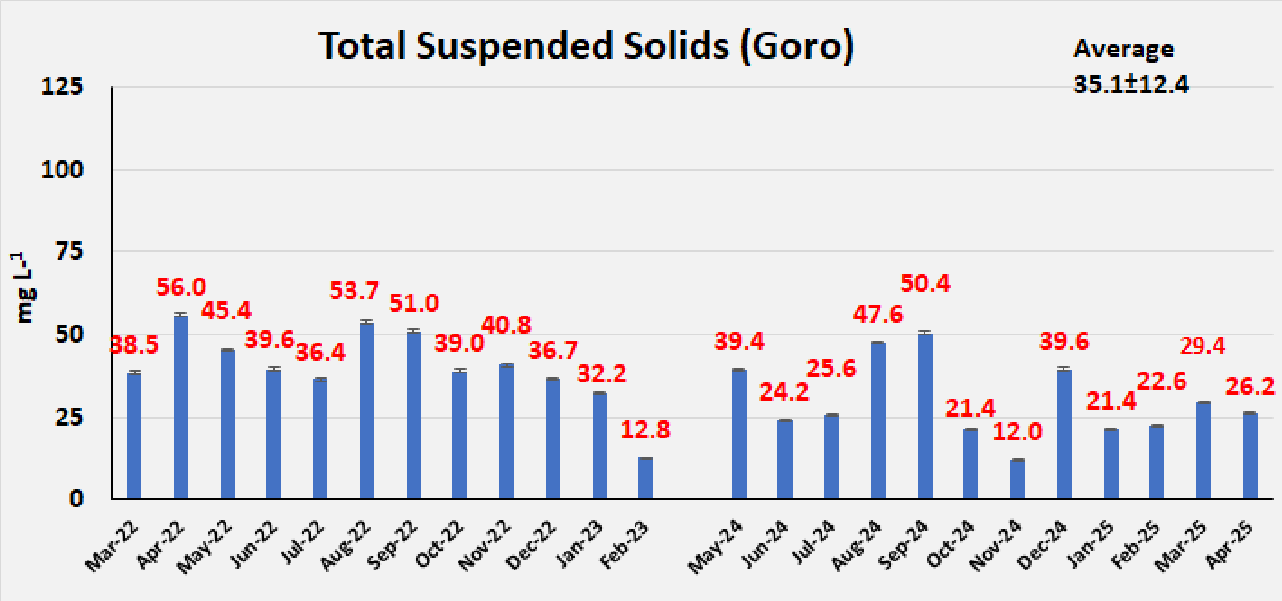




***Figure 11.*** *Variation of Dissolved Oxygen (DO) at Goro and Fattibello in the water column during the two sampling years.*

On average Total Suspended Solids (TSS) at Fattibello were slightly higher (41.4±24.0 mg L$^{-1}$) than at Goro (35.1±12.4 mg L$^{-1}$) with higher values in the first year (40.2±11.4 and 53.1±21.7 mg L$^{-1}$ at Goro and Fattibello respectively) in comparison to the second year (30.0±11.7 and 29.6±15.4 mg L$^{-1}$, respectively) in both stations (**Figure 8**).

At Goro the mean TSS monthly values ranged between 12.0 mg $L^{-1}$ in November 2024 and 56.0 mg $L^{-1}$ in April 2022, whereas at Fattibello they were between 13.4 mg $L^{-1}$ in January 2025 and 123.0 mg $L^{-1}$ in April 2022 (**Figure 12**) and fell within the *ex-ante* monitoring.



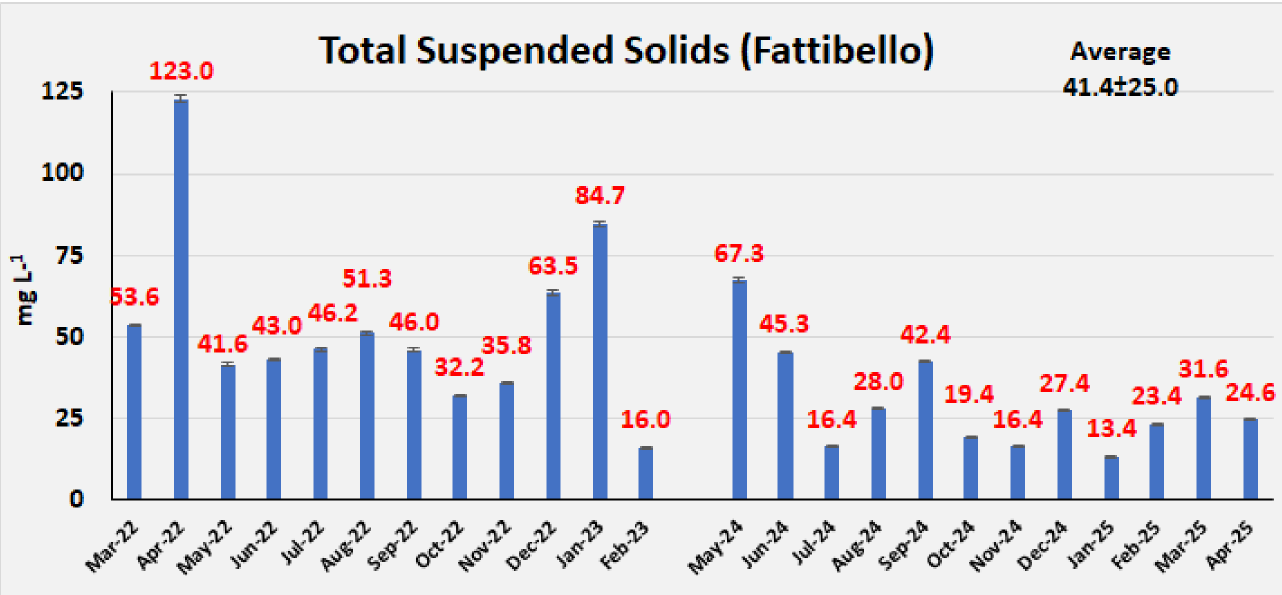




***Figure 12.*** *Variation of Total Suspended Solids (TSS) at Barbamarco and Caleri in the water column during the two sampling years.*

**Figures 13** and **Figure14** show the amount of settled particulate matter (SPM) collected in the sediment traps continuously between monthly samplings.

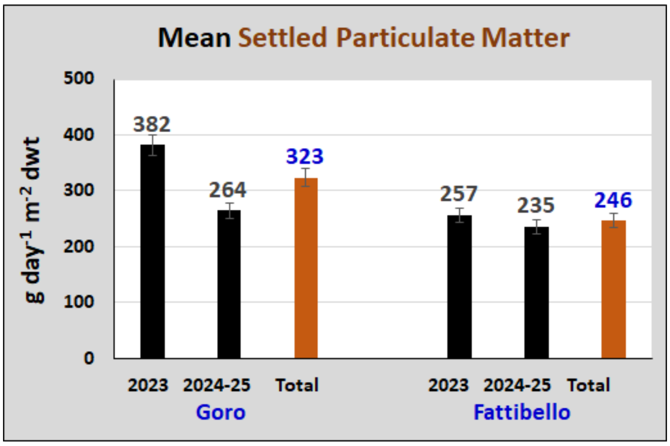



***Figure 13.*** *Mean values of Settled Particulate Matter (SPM) in the two stations.*

Goro showed an amount of SPM (323±246 g $m^{-2}$ $day^{-1}$) higher than Fattibello (246±174 g $m^{-2}$ $day^{-1}$) and the second years SPM were lower than the first year both at Goro (382±304 g $m^{-2}$ $day^{-1}$ in 2022-23; 264±163 in 2024-25) than at Fattibello (257±204 g $m^{-2}$ $day^{-1}$ in 2022-23; 235±146 in 2024-25).

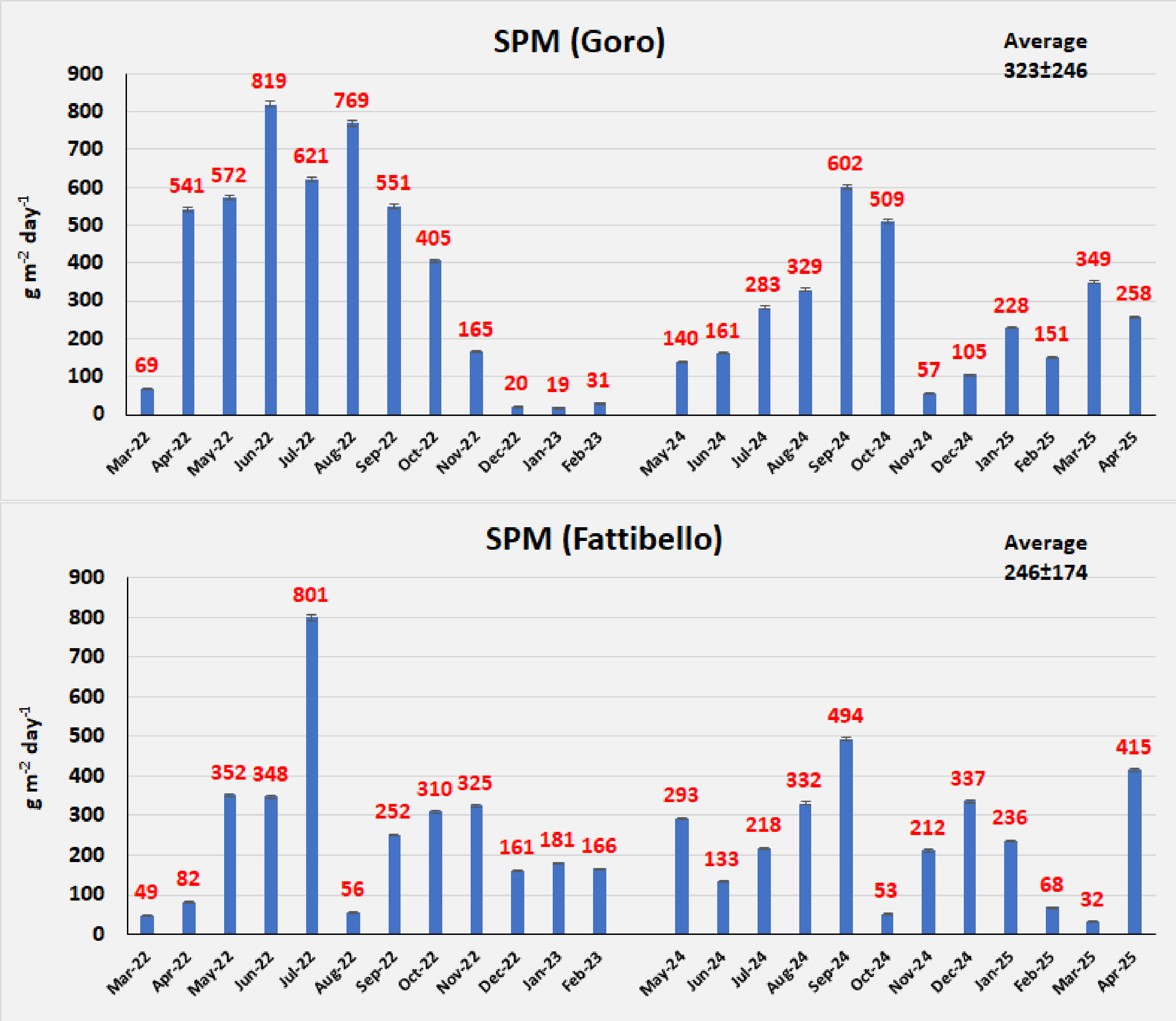



***Figure 14.*** *Variation of Settled Particulate Matter (SPM) at Goro and Fattibello in the water column during the two sampling years.*

The mean monthly SPM at Goro ranged between 19 g m$^{-2}$ day$^{-1}$ in January 2023 to 819 g m$^{-2}$ day$^{-1}$ in June 2022. At Fattibello SPM ranged between 32 g m$^{-2}$ day$^{-1}$ in March 2025 to 801 g m$^{-2}$ day$^{-1}$ in July 2022.

The mean concentration of nutrients in the water column (Reactive Phosphorus, Silicates, Ammonium, Nitrites, Nitrates, Dissolved Inorganic Nitrogen) is shown in **Figure 15**.

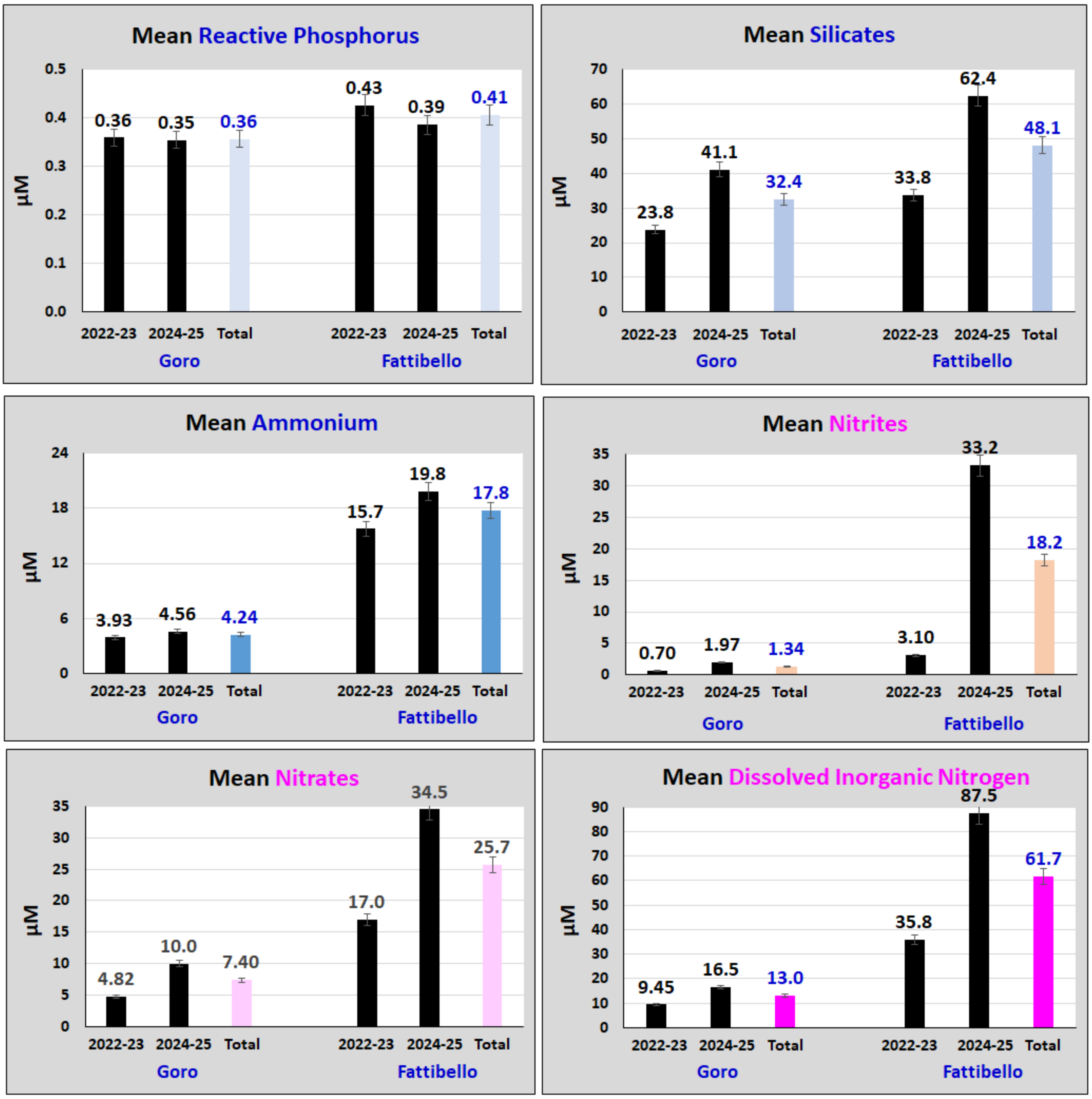
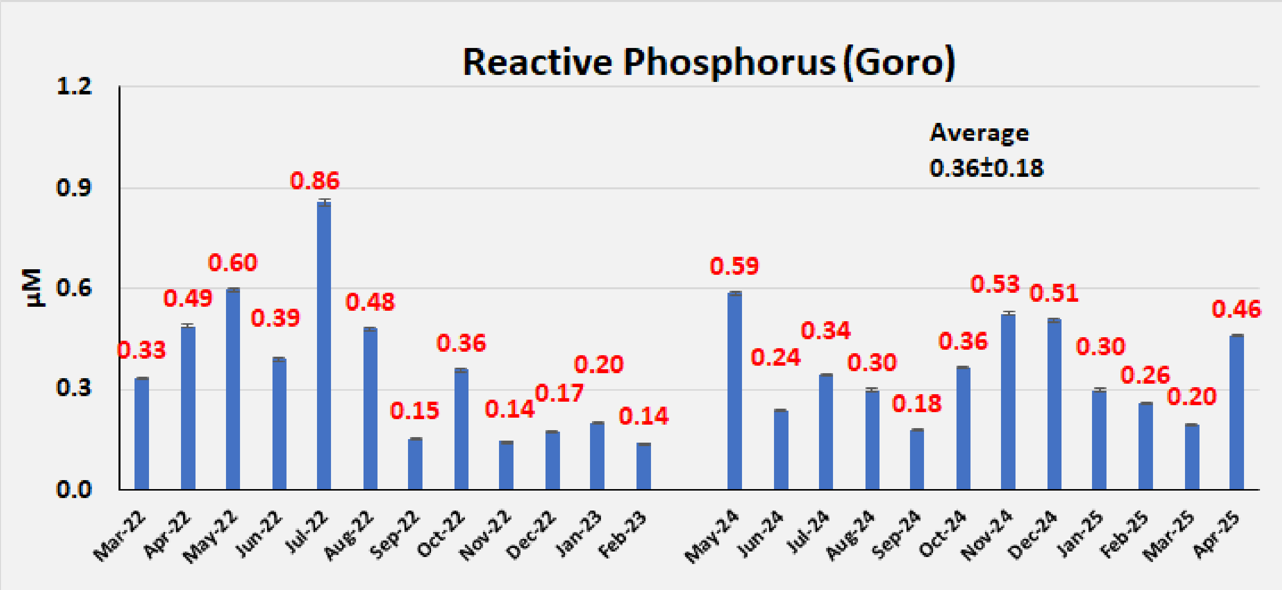



***Figure 15.*** *Variation of some nutrients in the water column in the Goro and Fattibello lagoons.*

The mean concentration of the Reactive Phosphorus (RP) at Goro (0.36±0.18 µM) was slightly lower than at Fattibello (0.41±0.23 µM) with negligible differences between the two years ranging from 0.36±0.22 µM and 0.35±0.14 µM at Goro and from 0.43±0.23 µM to 0.39±0.23 µM at Fattibello (**Figure 15**).



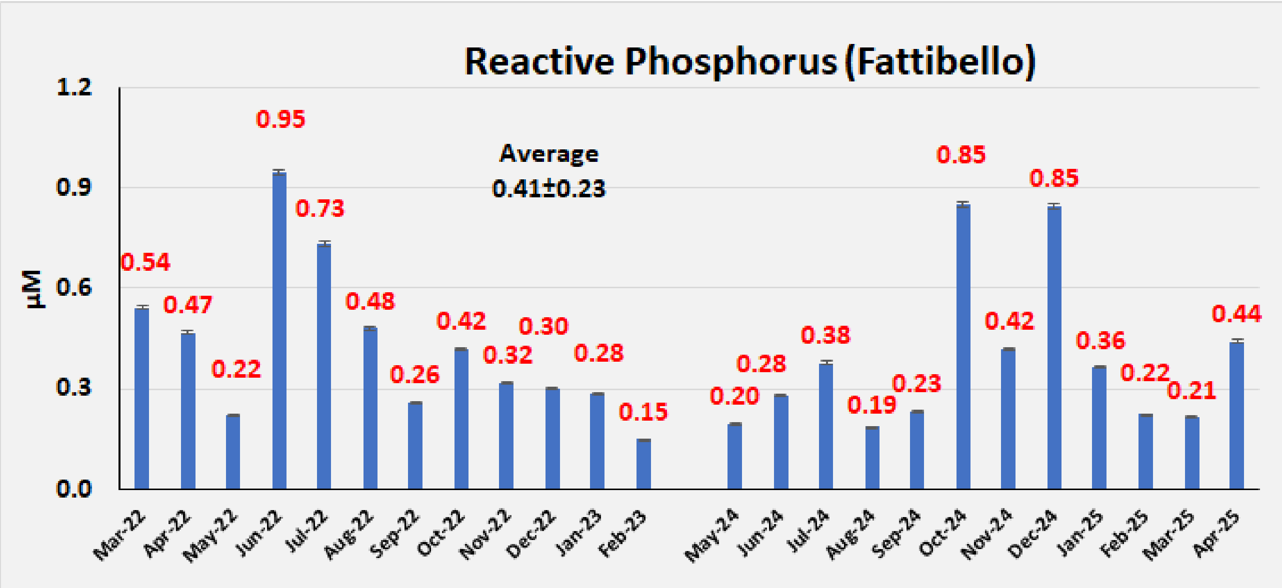
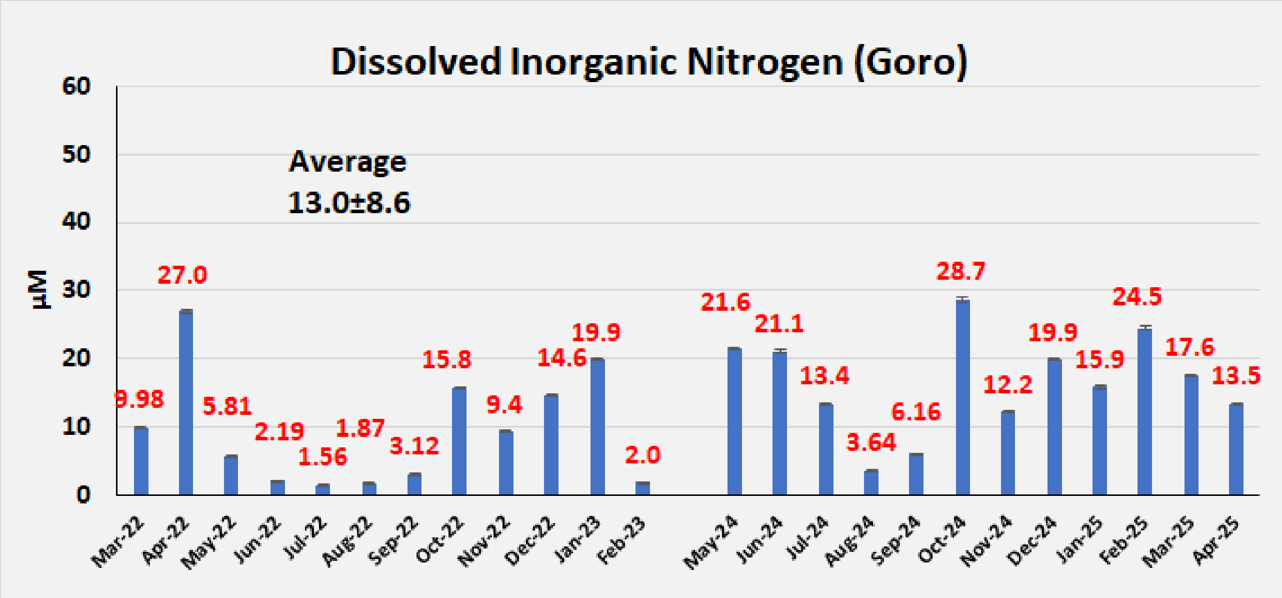




***Figure 16.*** *Variation of Reactive Phosphorus (RP) at Goro and Fattibello in the water column during the two sampling years.*

On monthly basis at Goro RP fluctuated between 0.14 µM in November 2022 and in February 2023 to 0.86 µM in July 2022. At Fattibello the lower value was 0.15 µM in February 2023 and 0.95 µM in January 2022 (**Figure 16**).

The mean Total Dissolved Inorganic Nitrogen (DIN) was almost five times higher at Fattibello (61.7±68.9 µM) than at Goro (13.0±8.6 µM). At Goro DIN ranged from 9.4±8.4 µM in 2022-23 to 16.5±7.3 µM in 2024-25. At Fattibello the mean DIN concentrations in the two different years were very different: 35.8±52.7 µM in 2022-23 and 87.5±75.5 µM in 2024-25.



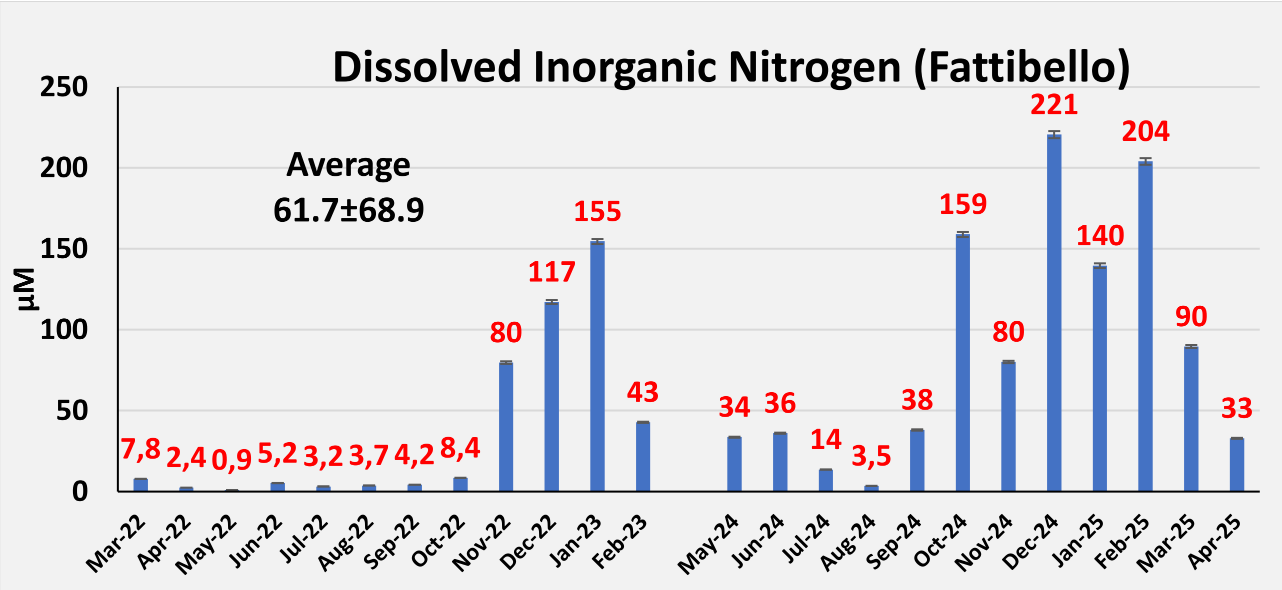




***Figure 17.*** *Variation of Dissolved Inorganic Nitrogen (DIN) at Goro and Fattibello in the water column during the two sampling years.*

On monthly basis, DIN at Goro ranged from 4.5 µM in June 2023 to 47.9 µM in December 2023 in October 2024 whereas at Fattibello DIN concentrations were between 0.9 µM in May 2022 to 221 µM in December 2024 (**Figure 17**).

If we consider the different species of DIN, i.e. ammonium, nitrites and nitrates (**Figure 15**), on average, the mean ammonium concentration at Goro (4.24 µM) was significantly lower than at Caleri (17.8 µM) with small changes in the two years. The *ex-ante* values were within these ranges.

Nitrites at Goro on average were 1.34 µM whereas at Fattibello, on average, they were particularly high (18.2 µM). Nitrates mean values ranged from 7.40 µM at Goro to 25.7 µM at Fattibello.

Similarly, the mean concentration of Silicates at Goro (32.4 µM) was lower than at Fattibello (48.1 µM) with significant changes in the two years.

The Phytoplankton as Chlorophyll-*a* concentrations and Macroalgal variables are shown in **Figure 18**.

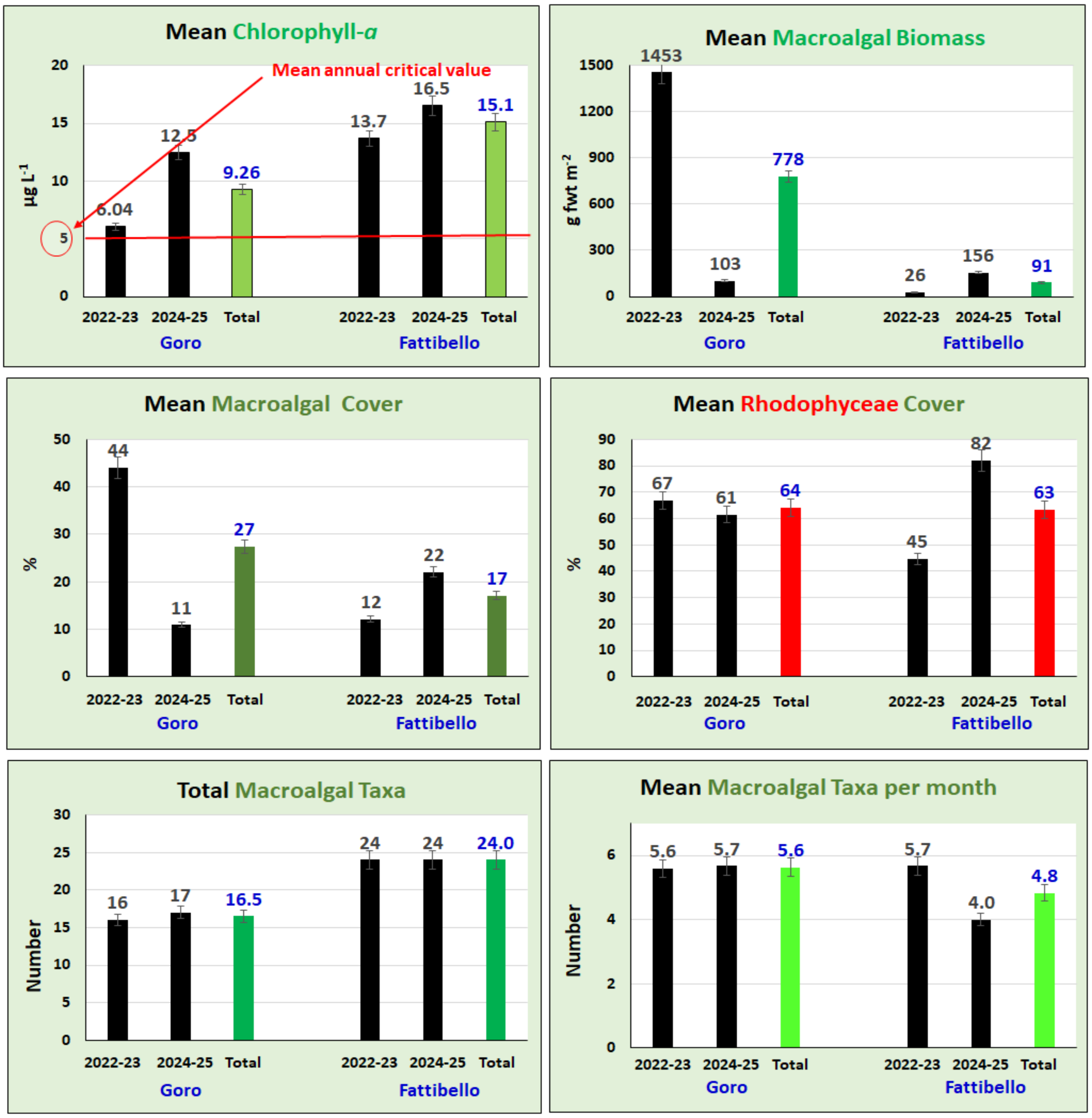



***Figure 18.*** *Variation of phytoplankton as total Chlorophyll-a and macroalgae at Goro and Fattibello.. The red line in the Chl-a concentrations is the mean limit that allow aquatic angiosperm rooting (Sfriso et al., 2023).*

On average, the mean concentration of total Chl-*a* both at Goro and Fattibello was markedly high: 9.26±8.23 µg $L^{-1}$ at Goro and 15.11±9.02 µg $L^{-1}$ at Fattibello (**Figure 18**), far exceeding the average limit concentration (2.5 µg $L^{-1}$) for the establishment of aquatic angiosperms found by Sfriso et al. (2023) by analyzing hundreds of data collected in more than 100 stations in Venice lagoon and in these Po Delta lagoon that correlate the presence/absence of aquatic angiosperms with the environmental parameters and in particular with the concentration of chlorophyll-*a*.

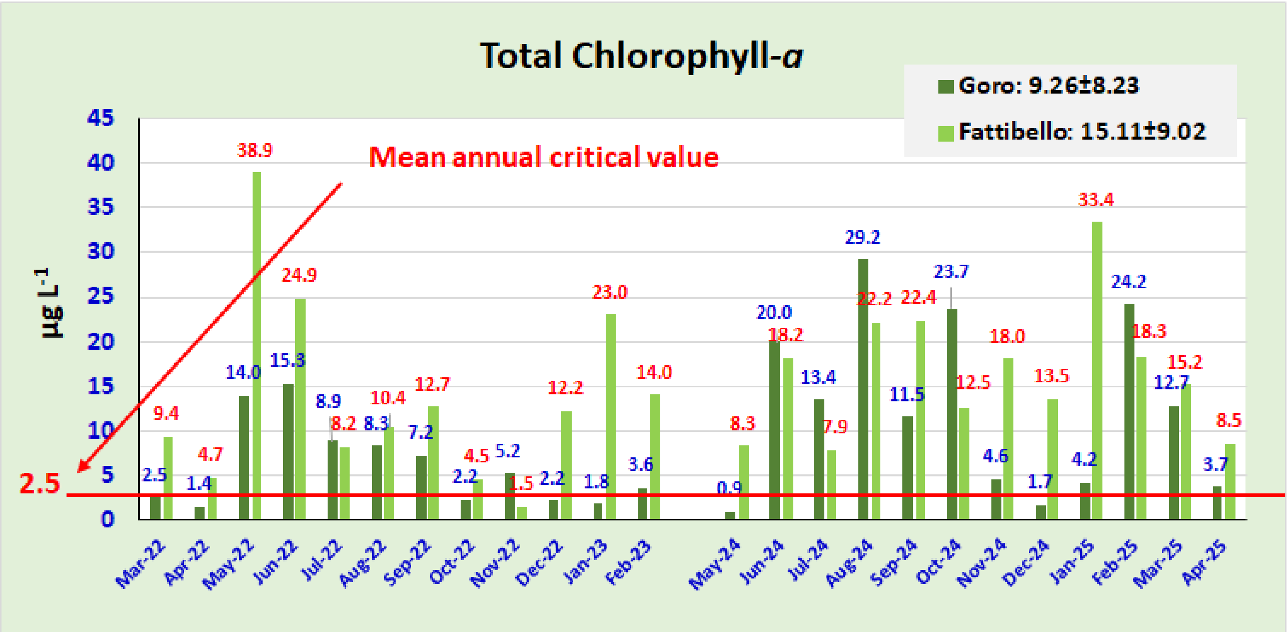



***Figure 19.*** *Variation of Total Chlorophyll-*a *at Goro and Fattibello in the water column during the two sampling years. The red line in the Chl-a concentrations is the mean limit that allow aquatic angiosperm rooting (Sfriso et al,, 2023).*

At Fattibello the Chlorophyll-*a* peak values where 38.9 µg $L^{-1}$ in May 2022 and 33.4 µg $L^{-1}$ in January 2025 whereas at Goro Chl-a peaked in August 2024 (29.2 µg $L^{-1}$) and February 2025 (24.2 µg $L^{-1}$) (**Figure 19**). Also Chl-*a* values recorded in these two years confirms the high values found in the *ex-ante* monitoring.

Conversely, macroalgal biomass was higher at Goro (778±1457 g fwt $m^{-2}$) than at Fattibello (91±144 g fwt $m^{-2}$) with important differences during the two years (**Figure 20**) due to the different locations of the stations during the two sampling years.

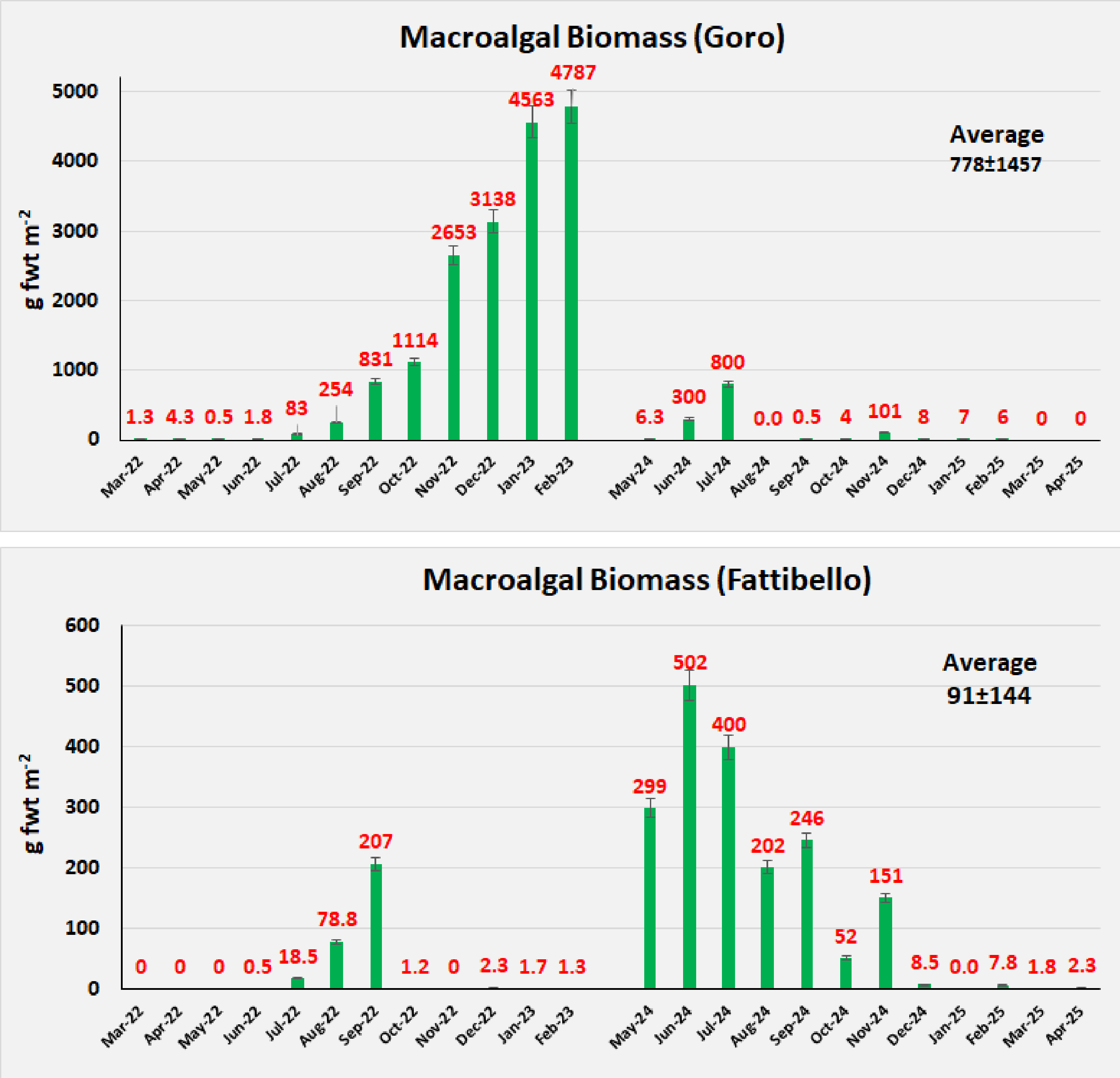



***Figure 20.*** *Variation of Macroalgal Biomass at Goro and Fattibello during the two sampling years.*

At Goro the average biomass decreased from 1453±1842 in 2022-23 to 103±11 g fwt m$^{-2}$ in 2024-25 but this was probably due to the different location of the sampling stations during the two years. At Fattibello the average biomass ranged from 26±61 in 2922-23 to 156±175 g fwt m$^{-2}$ in 2024-25. The monthly biomass trends in the two years is shown in **Figure 20**. At Goro in the first location the macroalgal biomass was high and from august 2022 to February 2023 progressively increased up to 4787 g fwt m$^{-2}$. The second year, in the new location, the biomass was significantly lower with a peak of 800 g fwt m$^{-2}$ in July 2024.

Rhodophyceae cover dominated on Chlorophyceae both at Goro (64%) and at Fattibello (63%). Rhodophyceae cover was similar at Goro in the two different years (67 and 61% in 2022-23 and 2024-25, respectively), whereas at Fattibello it increased from 45 to 82% (**Figure 18**).

The mean number of macroagal taxa recorded in the two stations was very hìgher than in the *ex-ante* because it is relative to 24 samplings. The number of taxa was 16.5 at Goro and 24.0 at Fattibello with negligible changes in the two sampling years. Similarly negligible differences were found in the number of taxa recorded every month at both stations: 5.6 at Barbamarco and 4.8 at Fattibello.

The total number of taxa recorded at Goro in the two years (24 monthly sampling days) was only 25 (14 Chlorophyceae, 10 Rhodophyceae and 1 Phaeophycea) (**Table 9**) and 9 taxa, representing most of the biomass, were alien taxa.

**Table 9.** Number of Taxa recorded at Goro. In pink the alien taxa.

| N° | N° | Chlorophyceae |
|---|---|---|
| 1 | 1 | *Blidingia dowsonii* (Hollenberg & I.A. Abbott) S.C. Lindstrom, L.A. Hanic & L . Golden |
| 2 | 2 | *Bryopsis hypnoides* J.V. Lamouroux |
| 3 | 3 | *Cladophora glomerata* (Linnaeus) Kützing |
| 4 | 4 | *Cladophora lehmanniana* (Lindenberg) Kützing |
| 5 | 5 | *Gayralia oxysperma* (Kützing) K.L. Vinogradova *ex* Scagel *et al.* |
| 6 | 6 | *Ulothrix implexa* (Kützing) Kützing |
| 7 | 7 | *Ulva australis* Areschoug |
| 8 | 8 | *Ulva compressa* Linnaeus |
| 9 | 9 | *Ulva rigida* C. Agardh |
| 10 | 10 | *Ulva intestinalis* Linnaeus |
| 11 | 11 | *Ulva polyclada* Kraft |
| 12 | 12 | *Ulva prolifera* O.F. Müller |
| 13 | 13 | *Ulva prolifera* subsp. *blidingiana* Alongi, Cormaci & Furnari |
| 14 | 14 | *Uronema marinum* Womersley |
| N° | N° | Rhodophyceae |
| 15 | 1 | *Acanthosiphonia echinata* (Harvey) Savoie & G.W.Saunders |
| 16 | 2 | *Agardhiella subulata* (C. Agardh) Kraft *et* M. J. Wynne |
| 17 | 3 | *Hypnea cervicornis* J Agardh |
| 18 | 4 | *Gracilariopsis longissima* (S.G. Gmelin) Steentoft *et al.* |
| 19 | 5 | *Gracilaria gracilis* (S.G. Gmelin) Steentoft *et al.* |
| 20 | 6 | *Gracilaria vermiculophylla* (Ohmi) Papenfuss |
| 21 | 7 | *Melanothamnus japonicus* (Harvey) Díaz-Tapia & Maggs |
| 22 | 8 | *Polysiphonia morrowii* Harvey |
| 23 | 9 | *Sahlingia subintegra* (Rosenvinge) Kornmann |
| 24 | 10 | *Solieria filiformis* (Kützing) P.W. Gabrielson |
| N° | N° | Phaeophyceae |
| 25 | 1 | *Kuckuckia spinosa* (Kützing) Kornmann |

**Table 10.** Number of Taxa recorded at Fattibello. In pink the alien taxa.

| N° | N° | Chlorophyceae |
|---|---|---|
| 1 | 1 | *Blidingia dowsonii* (Hollenberg & I.A. Abbott) S.C .Lindstrom, L.A. Hanic & L. Golden |
| 2 | 2 | *Blidingia ramifera* (Bliding) Garbary & L.B. Barkhouse |
| 3 | 3 | *Bryopsis cupressina* var. *adriatica* (J.Agardh) M.J. Wynne |
| 4 | 4 | *Chaetomorpha gracilis* Kützing |
| 5 | 5 | *Cladophora albida* (Nees) Kützing |
| 6 | 6 | *Cladophora dalmatica* Kützing |
| 7 | 7 | *Cladophora glomerata* (Linnaeus) Kützing |
| 8 | 8 | *Cladophora sericea* (Hudson) Kützing |
| 9 | 9 | *Cladophora vagabunda* (Linnaeus) C. Hoek |
| 10 | 10 | *Gayralia oxysperma* (Kützing) K.L.Vinogradova ex Scagel & al. |
| 11 | 11 | *Ulothrix flacca* (Dilllwyn) Thuret |
| 12 | 12 | *Ulva australis* Areschoug |
| 13 | 13 | *Ulva compressa* Linnaeus |
| 14 | 14 | *Ulva flexuosa* Wulfen |
| 15 | 15 | *Ulva linza* Linnaeus |
| 16 | 16 | *Ulva polyclada* Kraft |
| 17 | 17 | *Ulva prolifera* O.F. Müller |
| 18 | 18 | *Ulva prolifera* subsp. *blidingiana* Alongi, Cormaci & Furnari |
| 19 | 19 | *Ulva rigida* C. Agardh |
| 20 | 20 | *Uronema marinum* Womersley |
| | | **Rhodophyceae** |
| 21 | 1 | *Acrochaetium microscopicum* (Nägeli *ex* Kützing) Nägeli |
| 22 | 2 | *Agardhiella subulata* (C. Agardh) Kraft *et* M.J. Wynne |
| 23 | 3 | *Bostrychia scorpioides* (Hudson) Montagne |
| 24 | 4 | *Catenella caespitosa* (Withering) L.M. Irvine |
| 25 | 5 | *Caulacanthus okamurae Yamada* |
| 26 | 6 | *Dasya baillouviana* (S.G. Gmelin) Montagne |
| 27 | 7 | *Erythrotrichia carnea* (Dillwyn) J. Agardh |
| 28 | 8 | *Gracilariopsis longissima* (S.G. Gmelin) Steentoft *et al.* |
| 29 | 9 | *Gracilaria gracilis* (Stackhouse) Steentoft *et al.* |
| 30 | 10 | *Gracilaria vermiculophylla* (Ohmi) Papenfuss |
| 31 | 11 | *Melanothamnus japonicus* (Harvey) Díaz-Tapia & Maggs |
| 32 | 12 | *Polysiphonia morrowii* Harvey |
| 33 | 13 | *Polysyphonia* sp. |
| 34 | 14 | *Solieria filiformis* (Kützing) P.W. Gabrielson |
| | | **Phaeophyceae** |
| 35 | 1 | *Asperococcus ensiformis* (Delle Chiaje) M.J. Wynne |
| 36 | 2 | *Kuckuckia spinosa* (Kützing) Kornmann |
| 37 | 3 | *Myrionema orbiculare* J. Agardh |
| 38 | 4 | *Scytosiphon lomentaria* Link |

At Fattibello this number increased to 38 (20 Chlorophyceae, 14 Rhodophyceae and 4 Phaeophyceae) and also in this station the alien taxa with 8 species represented most of the biomass. In these two stations no sensitive taxa were recorded (**Table 10**).

A comparison with the number of taxa recorded in the *ex-ante* sampling is not possible because it refers to one sampling only.

**Surface sediments**

Some physico-chemical characteristics of the 5 cm sediment top layer in the two stations are reported in **Figure 21.**

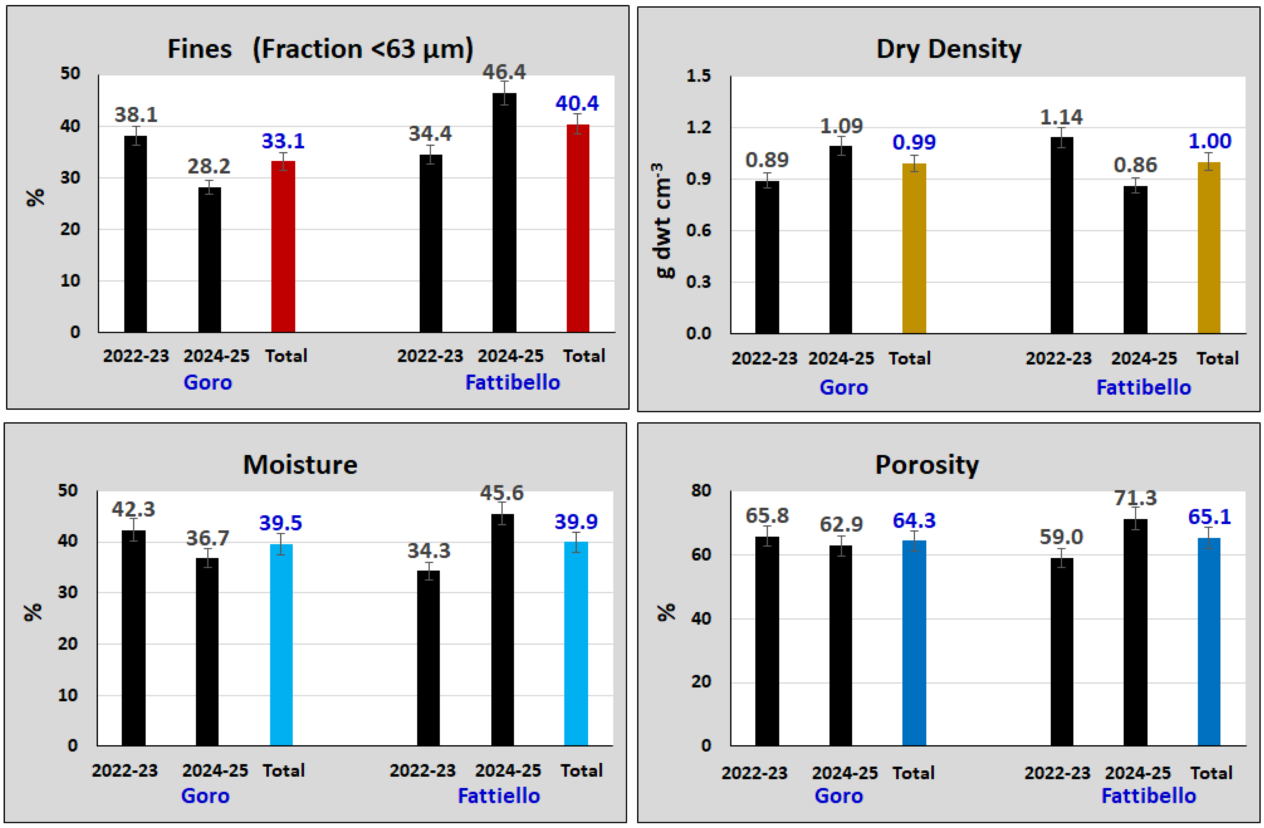



***Figure 21.*** *Variation of some physico-chemical characteristics of surface sediments*

Goro was a station with on average 33.1±16.2% of Fines, whereas at Fattibello Fines were on average 40.4±15.8 (**Figure 19**). The amount of dry sediment for volume unit (dry Density) in the two stations was quite similar (0.99±0.18 and 1.00±0.22 g dwt cm$^{-3}$, respectively) as well as Porosity (64.3±6.2 and 65.1±10.5%) and Moisture (39.5±6.0% and 39.9±9.0%).

Nutrient concentrations and carbon compounds in surface sediments are shown in **Figure 22**.

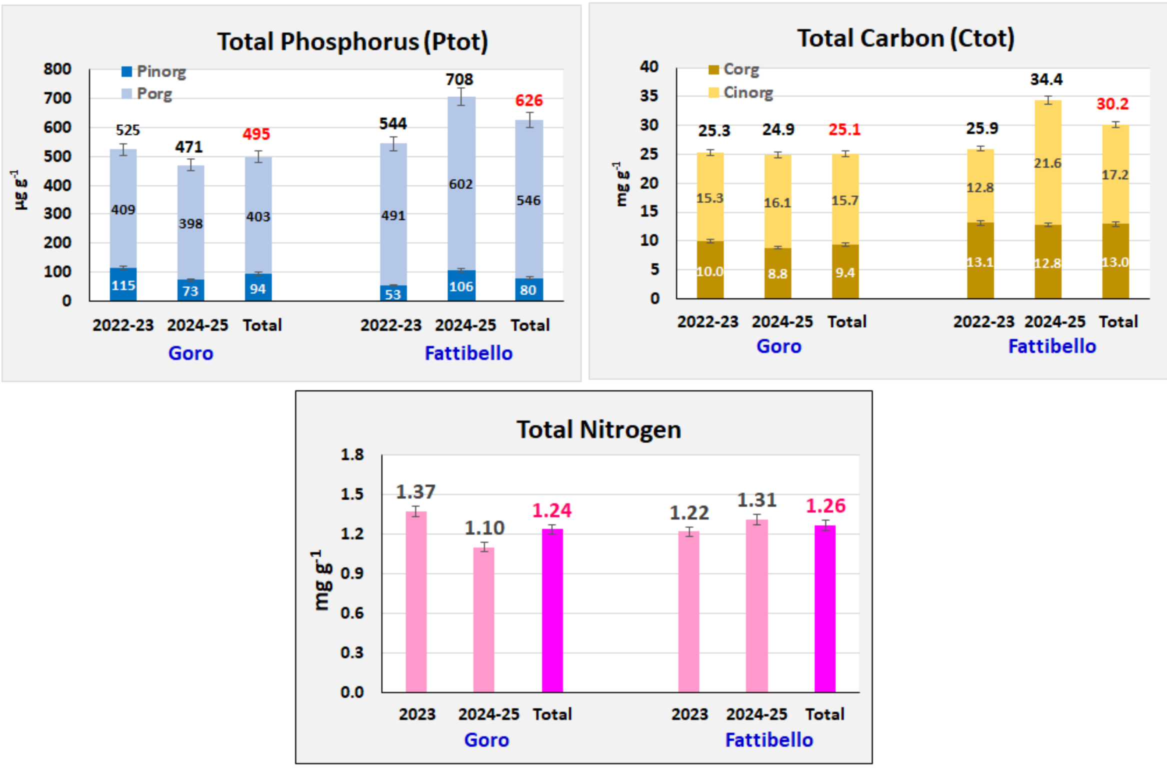



***Figure 22.*** *Average Phosphorus, Carbon and Nitrogen concentrations in surface sediments of the the two lagoons.*

The average Total Phosphorus (Ptot) concentration at Goro (495±126 μg $g^{-1}$) was slightly lower than at Fattibello (626±108 μg $g^{-1}$) (**Figures 22-23**). At Goro the concentration of the first year (525±98 μg $g^{-1}$) was higher than the second year (471±74 μg $g^{-1}$) whereas at Fattibello opposite results were obtained with a higher concentration (708±90 μg $g^{-1}$) than in the first year (544±43 μg $g^{-1}$). The inorganic fraction (Pinorg: 430±114 and 546±82 μg $g^{-1}$ at Goro and Fattibello, respectively) was significantly higher than the organic fraction (Porg: 65±37 and 80±49 μg $g^{-1}$ at Goro and Fattibello. At Goro the values were slightly higher because the station was moved from Val Seganda to Bassunsin (**Figure 4**).

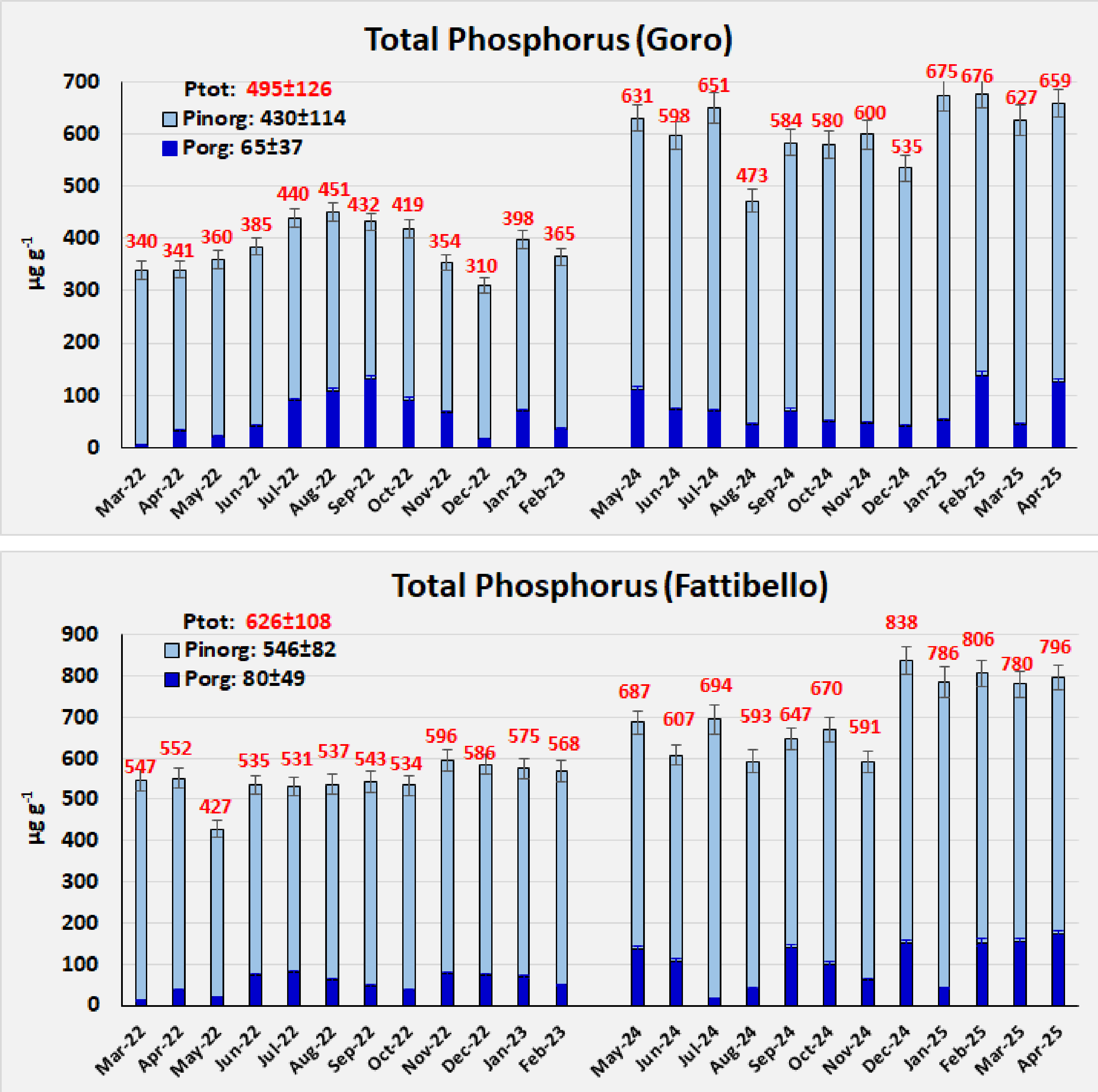



***Figure 23.*** *Variation of Organic Phosphorus (Porg), Inorganic Phosphorus (Pinorg) and Total Phosphorus (Ptot) in the top layer of surface sediments at Goro and Fattibello during the two sampling years.*

The trend of Total Nitrogen (Ntot) is reported in **Figures 22, 24**. The average concentrations recorded in the two years was quite similar (1.24±0.24 and 1.26±0.41 mg $g^{-1}$) with low differences during the two years. The highest concentrations were recorded at Fattibello (1.9 mg $g^{-1}$) in May 2024 and October 2024. At Goro the values were slightly higher because the station was moved from Val Seganda to Bassunsin (**Figure 5)**

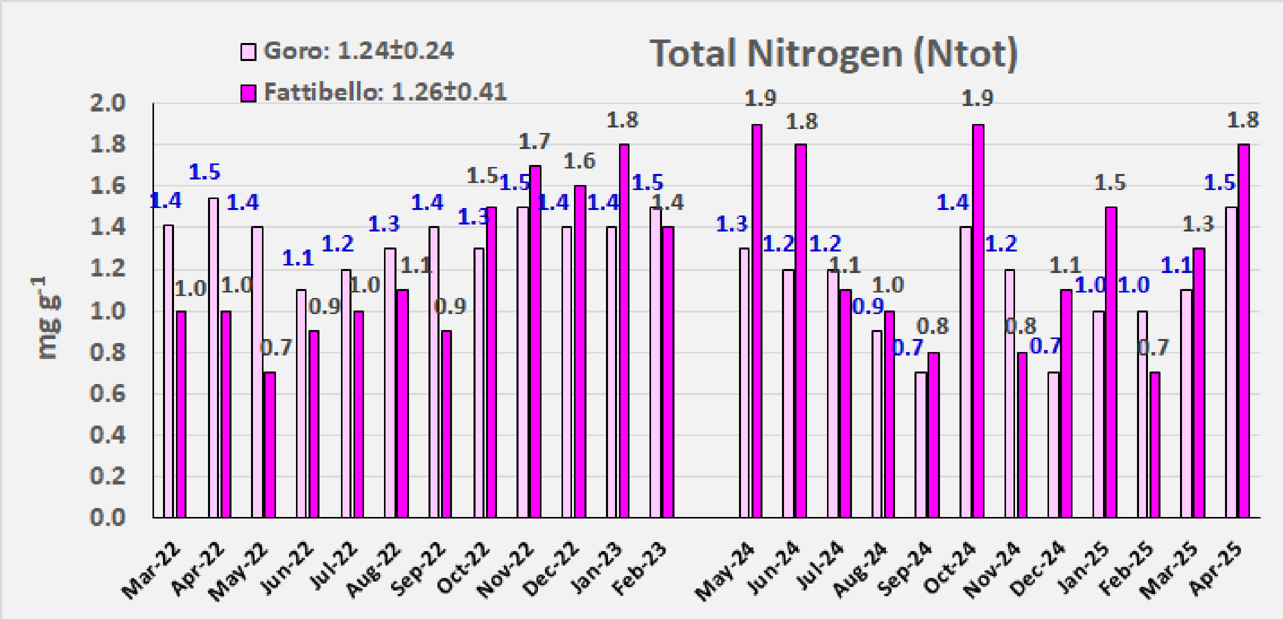
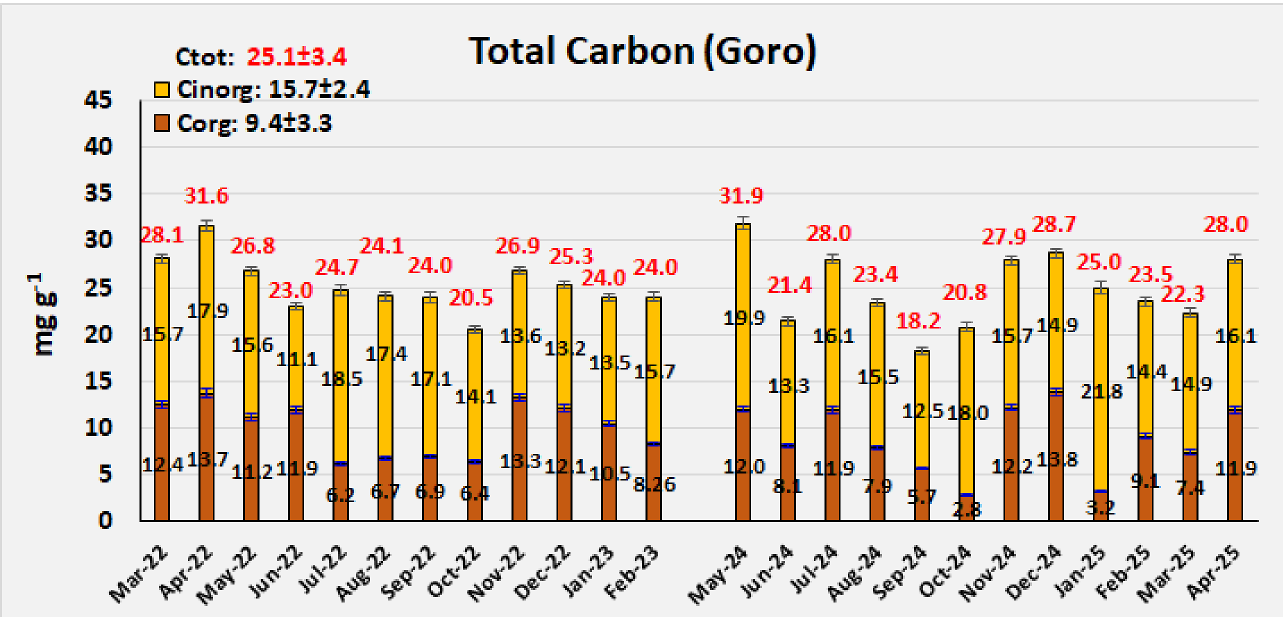



***Figure 24.*** *Variation of Total Nitrogen (Ntot) in the top layer of surface sediments at Goro and Fattibello during the two sampling years.*

Total Carbon (Ctot) at Goro on average was lower (25.1±3.4 mg $g^{-1}$) than at Fattibello (30.2±5.7 mg $g^{-1}$) and the inorganic fraction (Cinorg) was slightly higher than the organic one (Corg) (**Figures 22, 25**). At Goro Ctot concentrations ranged between 18.2 in September 2024 to 31.9 in May 2024 whereas at Fattibello Ctot was between 20.4 June 2022 and 41.1 in October 2024 (**Figure 23**). At Goro the values were slightly higher because the station was moved from Val Seganda to Bassunsin (**Figure 5).**



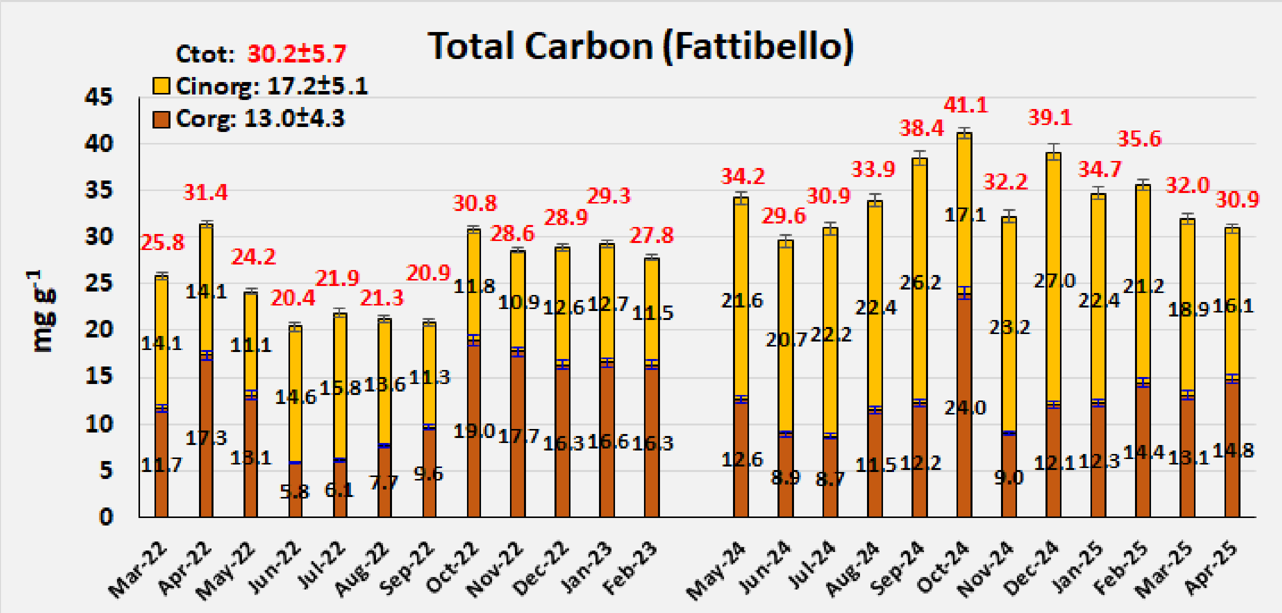



***Figure 25.*** *Variation of Organic Carbon (Corg), Inorganic Carbon (Cinorg) and Total Carbon (Ctot) in the top layer of surface sediments at Goro and Fattibello during the two sampling years.*

# SECTION 2 – Ecological Quality Status

## Macrophytes and MaQI determination

***Methods***

***Macrophyte coverage and taxa determination.***

Macroalgae samples were collected in accordance with the method for the determination of the Macrophyte Quality Index (MaQI, ISPRA, 2011; Sfriso et al., 2014, **Figure 26**) in order to determine the ecological status as required by Water Framework Directive (2000/60/EC).

**Macrophyte Quality Index (MaQI)**

| | Macroalgae (score): Opportunistic 0 | Indifferent 1 | Sensitive 2 | Ecological Quality Ratio (EQR) | | | |
|---|---|---|---|---|---|---|---|
| Macroalgae | Every coverage (1) | | ≥25% | 0.85 | | | 1 |
| | | | 15-25% | 0.65 | | 0.75 | 0.85 |
| | | | ≤15% | 0.55 | 0.55 | 0.65 | |
| | Total coverage ≤5% | | 2 species | 0.45 | | | |
| | Total coverage >5% | Rhodophyta dominance | ≤2 species | 0.35 | | | |
| | | Chlorophyta dominance | ≤2 species | 0.25 | | | |
| | Total coverage ≤5% | | 1 | | | | |
| | | | 0 | 0.15 | | | |
| | Absent/Trace (2) | | | 0 | | | |
| aquatic angiosperms | *Ruppia cirrhosa, R. maritima, Zostera noltii* | | | missing | <50% (3) | 50-75% | >75% |
| | *Zostera marina* | | | | <25% | 25-75% | >75% |
| | *Cymodocea nodosa* | | | missing | | <25% | ≥25% |
| | *Posidonia oceanica* | | | missing | | | Present |

| | |
|---|---|
| (1) | Per cent species number. |
| (2) | The Xanthophycea: *Vaucheria* spp. can be present with a coverage up to 100%. Seasonal growth of Rhodophyta and/or Chlorophyceae which are not able to bloom. |
| (3) | Per cent species coverage |

***Figure 26**. MaQI Scheme*

At each station, the relative coverage of macroalgae was assessed by using the Visual Census Technique in clear waters, or touching the bottom 20 times with a rake in turbid waters, in order to discriminate a coverage ≥5%, as required by the application of the index. Subsequently 5-6 macroalgal samples were collected reporting the percentage of

Chlorophyta and Rhodophyta. Representative samples of all the species present in the stations were stored in 4% formaldehyde for the taxonomic determination.

**Results**

The application of the Macrophyte Quality Index (MaQI) from 2021 (*ex-ante*) to 2025 is shown in **Table 11**. The *ex-ante* sampling shows an Ecological Quality Ratio (EQR = 0.25) accounting for Poor conditions. Goro in the *ex-ante* sampling carried out in Seganda site (**Figure 4)** was covered by a significant biomass of *Ulva australis* (Chlorophyceae). After a first transplant tentative, due to the *Ulva* biomass and high-water turbidity which did not allow the rooting of the first transplants the station was moved to the Bassunsin site where the water was clearer and the macroalgal biomass was mainly represented by Rhodophyceae. In 2023 and 2024 no angiosperm rooting was observed and the EQR was 0.35 accounting for Poor conditions. In autumn 2025 after the continuous transplanting efforts carried out by fishermen and DAIS-UNIVE staff, some small, scattered patches of *Nanozostera noltei* were formed, indicating a first rooting success which we hope will increase over time, forming more developed patches and small meadows. In 2025 the EQR was 0.55 accounting for Moderate conditions.

In the *ex-ante* sampling Fattibello site was dominated by the growth of *Ulva australis*. Under these conditions the EQR was 0.25 accounting for Poor conditions. Therefore, in 2023 transplants were carried out in a nearby area where the macroalgal biomass was negligible. However, in 2023 and 2024 the high-water turbidity hampered the aquatic angiosperm rooting. In autumn 2025 some tufts of *Ruppia cirrhosa* were recorded but without the appearance of additional tufts due to the expansion of the species. Therefore, the EQR remained 0.35 (Poor conditions). However, if in 2026 these tufts will show a consolidated rooting with their expansion the EQR will be 0.55 (Moderate conditions)

***Table 11**. Application of the Macrophyte Qualitry Index (MaQI) based on two sampling dates: one in spring (May) and the other in autumn (November) as required by the normative (2000/60/EC).*

| Station | Year | Ecological Qualty Ratio (EQR) | Ecological status |
|---|---|---|---|
| Goro | 2022 (*ex ante*) | 0.25 | Poor |
| | 2023 | 0.35 | Poor |
| | 2024 | 0.35 | Poor |
| | 2025 | 0.55 | Moderate |
| Fattibello | 2022 (*ex ante*) | 0.25 | Poor |
| | 2023 | 0.35 | Poor |
| | 2024 | 0.35 | Poor |
| | 2025 | 0.35 | Poor |

In both stations no high-quality macroalgal species (sensitive species) were recorded either in the *ex-ante* sampling or in the following years. However, the presence of aquatic angiosperms and their diffusion, improving ecological conditions, will certainly allow the growth of sensitive algal species as is happening in the Venice lagoon after the transplants carried out within the Life SERESTO project (Life12 NAT/IT/000331) (Sfriso et al., 2021) However, in October 2025, while this report was being written, tufts of *Nanozostera noltei* were observed at the first-year transplant station, which bodes well for the spread of this species in the coming years, as has already happened in the Venetian Lagoon, where two-thirds of the colonization occurred between the fifth and tenth year after the first transplants.

**Statistical analyses**

The principal component analysis (PCA) of all the data collected in the two stations sampled monthly during 2022-23 and 2024-25 periods (48 samplings) shows 10 components explaining a total variance of 80.8%. Significant values (loading >0.7) are shown by the first two components (variance 35.1%) which are plotted in a plane in **Figure 27**.

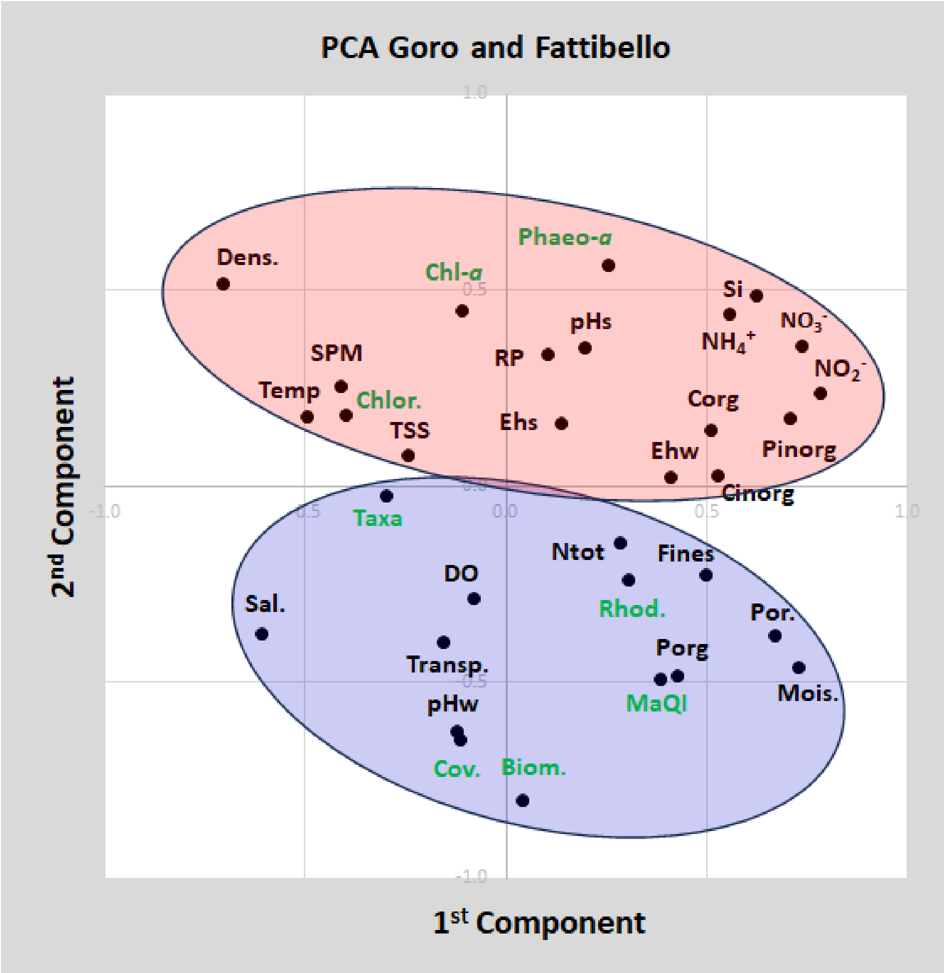



***Figure 27.*** *PCA analysis between environmental parameters and macrophyte variables recorded at Goro and Fattibello*

*Legenda.* ***Macrophyte variables****: Ang. (Aquatic angiosperms), Biom. (Macroalgal biomass), Chl-a (Chlorophyll-a), Chlor. (Chlorophyceae abundance), Cov. (Macroalgal cover), MaQI (Macrophyte Quality Index), Phaeo-a (Phaeophytin-a), Rhod. (Rhodophyceae abundance), Taxa (Number of macroalgae).* ***Environmental parameters:*** *Ammonium ($NH_4^+$), Density (Dens.), Inorganic Carbon (Cinorg), Inorganic Phosphorus (Pinorg), Nitrites ($NO_2^-$), Nitrates ($NO_3^-$), Moisture (Moist.), Organic Carbon (Corg), Organic phosphorus (Porg.), Porosity (Por.), Reactive Phosphorus (RP), Salinity (Sal.), Sediment Eh (Ehs), Sediment fraction <63 µm (Fines), Sediment pH (pHs), Settled Particulate Matter (SPM), Silicates (Si), Temperature (Temp.), Transparency (Transp.), Total Nitrogen (Ntot), Total Suspended Solids (TSS): Water Eh (Ehw), Water pH (pHw).*

They are characterized by the significant (p> ±0.7) negative values of the macrophyte variables: Macroalgal biomass (-0.804) and surface sediment Density (-0.70). The positive contribute was due to the water column Nitrites (-0.78) and Nitrates (-0.74) and the surface sediment Moisture (0.73) and Pinorg (0.71).

Two distinct groups can be visualized: one on the top side, indicating the worst conditions characterized by Chlorophyll-*a*, Phaeophytin-*a* and Chlorophyceae associated with water parameters: i.e. Temperature, SPM, TSS, Ammonium, Nitrites, Nitrates, Reactive

Phosphorus, Silicates, Ehw and some sediment parameters: i.e. Density, pHs and Ehs, Pinorg, Cinorg and Corg. The other, on the bottom side, indicating the better environmental conditions, was characterized by MaQI, the number of taxa, Rhodophyceae abundance, Macroalgal biomass and Macroalgal cover associated with Salinity, water Transparency, pHw and the main sediment characteristics: Fines, Moisture, Porosity, Total Nitrogen and Organic Phosphorus, Organic and Inorganic Carbon, Organic and Inorganic Phosphorus and Total Nitrogen.

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## Macrobenthos and M-AMBI and BITS determination

### *Methods*

The macrobenthic community analysis (composition, structure and dynamics) is the best approach for assessing the ecological status for a given water body. The DL 260 / 10 explicitly requests the analysis of the "macrobenthos" as biological quality element for the definition of the ecological status of transition and marine-coastal waters. The monitoring of the macrobenthic community on a spatio-temporal scale enables the evaluation of the effectiveness of activities carried out for environmental improvement and it provides an adequate tool for a rigorous assessment of the quality of the environmental conditions.

The macrobenthos monitoring actions are carried out at 2 sites of the Natura 2000 Network, characterized by the presence of the priority habitat 1150 - coastal lagoons, and affected by the conservation activity of this habitat by transplanting marine phanerogams; at each of the 4 sites (Sacca di Goro, Valle Fattibello,), the monitoring action is carried out at several stations. Initially the Seganda station was included in Goro, but following the unsatisfactory results obtained during the first 2 years of experimentation it was decided to abandon it as the turbidity of the water is excessive all year round for the development of macrophytes; in Goro the experimentation is therefore continued in Bassunsin. The same negative conditions also manifested themselves in the station initially chosen in Fattibello; here, after a short time, a colony of flamingos settled, whose activity makes the water constantly cloudy. for this reason the transplant site, after 2 years, was moved to another area of the lagoon, away from the flamingos.

At each station, macrobenthos is sampled following a BACI (Before-After, Control-Impact) design, that is sampling in a control site and in a site subjected to impact (specifically the transplant operation), before and after impact (intervention) (**Fig. 1**).

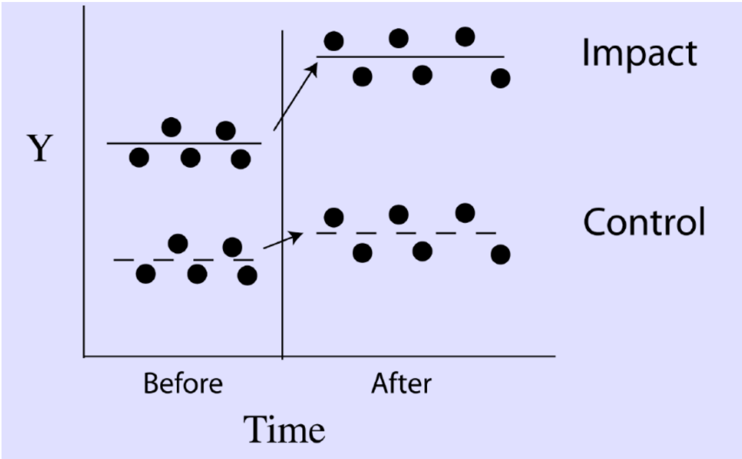



*Figure 1. Before-after-control-impact (BACI) design*

At each station 5 replicates of samples are collected in order to achieve for each site (i.e. lagoon) 5 Control replicates, i.e. in areas not directly interested by the phanerogam transplant; 5 Impact replicates, i.e. in areas directly interested by the phanerogam transplant.

For the assessment of environmental quality, an indicator specifically required by Legislative Decree 260 / 2010 for transitional waters is used, the M-AMBI index (Muxika et al., 2007).

After the ex-ante campaign (May 2021), and the beginning of phanerogams transplant operations (autumn 2021), sampling of macrobenthos was carried out twice a year, i.e. in April and October 2022, May and November 2023, and the most recent April 2024. Samples from April 2024 are under analyses, so will be not included in this report.

Structural indices describing the macrobenthic community were calculated on species/abundance data at each replicate of each station.

The ecological quality at each station has been assessed by applying the macrobenthic index M-AMBI (Muxika et al., 2007) on the species/abundance data set. The M-AMBI index is based on a multivariate analysis in which factor analysis combines the values of AMBI, with those of Shannon-Wiener diversity (H') and number of species (S). The M-AMBI is calculated by means a user-friendly software (www.azti.es) to be applied with the latest update of the species list already available.

The index is based on the classification of macrobenthic species into 5 ecological groups (EG) which correspond to different levels of disturbance-sensitivity (Borja et al., 2000). The

EGI group includes the most sensitive species; following a tolerance gradient we arrive at the EGV group, which includes strongly opportunistic species, characteristic of heavily polluted environments. The AMBI index is calculated as:

AMBI = [(0*%EGI)+(1,5*%EGII)+(3*%EGIII)+(4,5*%EGIV)+(6*%EGV)]/100

The ecological value (EG) of benthic taxa is reported in the AMBI library. If some species are not assigned an ecological value, as such species are not present in the AMBI library, the accuracy of the result may be compromised if: a) the percentage of unassigned taxa is > 20%, b) the taxa not belonging to some groups have a large number of individuals. Thanks to the calculation method, the M-AMBI Index is able to summarize the complexity of soft bottom communities, enabling the ecological reading of the ecosystem in question. M-AMBI corrects the quality values provided by AMBI through the integration of diversity and specific richness. M-AMBI is an extremely flexible tool for the derivation of the EQR, as it requires the operator to enter the limit values (equivalent to the reference values) for H ', S and AMBI. If this step is omitted, the reference values for the "High" quality class are taken as the highest values of S and H '(and lowest of AMBI) present within the numeric matrix for which the operator is running the calculation. This omission leads to extreme errors in the evaluation of the EQ (ecological quality).

The value of the M-AMBI varies between 0 and 1. Below (**Fig. 2**) are reported (i) the type-specific reference values for each metric that makes up the M-AMBI, the M-AMBI class limits, expressed in terms of the ecological quality ratio (RQE), between the High status and the Good status, and between the Good status and the Moderate status as required by current legislation. The values of the reference conditions and the relative Good / Moderate and High / Good limits considered for the calculation are those relating to macrotypes 1 and 2 (M-AT-1, M-AT-2), to which Fattibello and the Sacca di Goro respectively belong.

*Valori di riferimento e limiti di classe*

***Tab. 4.4.1/c – Limiti di classe in termini di RQE per l'M-AMBI***

| *Rapporto di Qualità Ecologica* | | | |
|---|---|---|---|
| *Elevato/Buono* | *Buono/Sufficiente* | *Sufficiente/Scarso* | *Scarso/Cattivo* |
| 0,96 | 0,71 | 0,57 | 0,46 |

Le condizioni di riferimento sono state definite sulla base di un criterio misto statistico/geografico. L'indice M-AMBI è un indice multivariato, pertanto le condizioni di riferimento vanno indicate per i tre indici che lo compongono: AMBI, Indice di Diversità di Shannon-Wiener e numero di specie (S).

***Tab. 4.4.1/d - Valori di riferimento tipo-specifiche per l'applicazione dell'M-AMBI***

| Macrotipo | Geomorfologia | Escursione marea | Salinità | AMBI | Diversità di Shannon-Wiener | Numero di Specie (S) |
|---|---|---|---|---|---|---|
| M-AT-1 | Laguna costiera | Non tidale | - | 1,85 | 3,3 | 25 |
| M-AT-2 | Laguna costiera | microtidale | Oligo/meso/poli | 2,14 | 3,40 | 28 |
| M-AT-3 | Laguna costiera | microtidale | Eu/iper | 0,63 | 4,23 | 46 |

*Fig. 2. Reference values for the M-AMBI calculation (from the DL260/10).*

The BITS index (Mistri & Munari, 2008) was also applied. BITS is written:

$$BITS = \log[(6fI+fII) / (fIII+1)+1] + \log[nI/(nII+1) + nI/(nIII+1) + 0{,}5nII/(nIII+1) +1]$$

where *fI* is the sensitive families frequency (ratio of the total number of individuals belonging to sensitive families to the total number of individuals in the sample), *fII* is the tolerant families frequency (ratio of the total number of individuals belonging to tolerant families to the total number of individuals in the sample), and *fIII* is the opportunistic families frequency (ratio of the total number of individuals belonging to opportunistic families to the total number of individuals in the sample). The +1 terms in the equation are needed in order to allow the division operation to be completed even when *fIII* is null, and to prevent the eventuality of a log of zero if *fI* and *fII* are null. The second term of the BITS model allows to weight the number of sensitive families respect the tolerant and the opportunistic ones: *nI* is the number of sensitive families, *nII* is the number of tolerant and *nIII* is the number of opportunistic families. Again, the +1 terms in the equation are needed in order to allow the division operation to be completed. The BITS index is null when there are no sensitive and tolerant families, indicating a very high amount of organic matter in the sediments, and, in lagoonal ecosystems, a very poor water exchange. BITS is high when the environment is good, with few opportunistic families, and it decreases as the environment degrades.

The following scheme (from DL260/10) reports the BITS class limits, expressed in terms of the ecological quality ratio (RQE), between the High status and the Good status, and between the Good status and the Moderate status as required by current legislation, and the type-specific reference values for BITS.

| **Limiti di classe in termini di RQE per il BITS** | | | |
|---|---|---|---|
| Elevato/Buono | Buono/Sufficiente | Sufficiente/Scarso | Scarso/Cattivo |
| 0,87 | 0,68 | 0,44 | 0,25 |

| **Valori di riferimento tipo-specifiche per l'applicazione del BITS** | |
|---|---|
| M-AT-1 | 2,8 |
| M-AT-2 | 3,4 |
| M-AT-3 | 3,4 |

***Results***

**Sacca di Goro lagoon**

Overall, a total of 37 macrobenthic taxa were found in the Sacca di Goro. **Table 1** shows the faunal list of the macrobenthic taxa collected during all the monitoring campaigns. Relative abundances vary according to season and site, and the variations are better highlighted below in the analysis of community structural indices

| | |
|---|---|
| *Alitta succinea* | *Gammarus aequicauda* |
| *Capitella capitata* | *Gammarus crinicornis* |
| *Capitella minima* | *Gammarus insensibilis* |
| *Fabricia sp* | *Gastrosaccus sanctus* |
| *Fabricia stellaris* | *Grandidierella japonica* |
| *Hediste diversicolor* | *Grandidierella sp* |
| *Heteromastus filiformis* | *Idotea balthica* |
| *Janua pagenstecheri* | *Melita palmata* |
| *Mediomastus fragilis* | *Microdeutopus gryllotalpa* |
| *Oligochaeta* | *Monocorophium acherusicum* |
| *Polydora cornuta* | *Monocorophium insidiosum* |
| *Pygospio elegans* | *Arcuatula senhousia* |
| *Spio filicornis* | *Hydrobia acuta* |
| *Streblospio eridani* | *Hydrobia ventrosa* |
| *Streblospio shrubsolii* | *Lentidium mediterraneum* |
| *Carcinus maenas* | *Ruditapes philippinarum* |
| *Chironomus salinarius* | *Actinia spp* |
| *Corophium orientale* | *Diadumene sp* |
| *Cyathura carinata* | |

*Tab. 1. Taxonomic list of the macrobenthic taxa identified in the Sacca di Goro*

In **Table 2** the values of community descriptors (diversity, H’, and species richness, S), together with AMBI/M-AMBI values and ES are reported for all the sampling dates. In the Table also the reference parameters are shown (in grey).

| *Date* | *Stations* | *AMBI* | *H'* | *S* | *M-AMBI* | *Status* |
|---|---|---|---|---|---|---|
| | Bad | 6 | 0 | 0 | 0 | Bad |
| | High | 2.14 | 3.4 | 28 | 1 | High |
| June 2021 | Gor-C | 2.98 | 2.71 | 15 | 0.71 | Moderate |
| | GorS | 2.90 | 2.47 | 23 | 0.78 | Good |
| April 2022 | Gor-C | 3.13 | 2.25 | 20 | 0.71 | Moderate |
| | GorS-Tr | 3.31 | 2.64 | 16 | 0.68 | Moderate |
| | GorB-Tr | 2.9 | 2.18 | 19 | 0.71 | Moderate |

| | | | | | | |
|---|---|---|---|---|---|---|
| October 2022 | Gor-C | 4.24 | 1.41 | 14 | 0.46 | Bad |
| | GorS-Tr | 3.13 | 1.47 | 7 | 0.48 | Poor |
| | GorB-Tr | 2.63 | 3.11 | 21 | 0.85 | Good |
| May 2023 | Gor-C | 3.08 | 2.67 | 15 | 0.68 | Moderate |
| | GorS-Tr | 3.54 | 2.70 | 18 | 0.69 | Moderate |
| | GorB-Tr | 1.73 | 1.49 | 21 | 0.76 | Good |
| November 2023 | Gor-C | 2.98 | 1.96 | 9 | 0.56 | Poor |
| | GorS-Tr | 3.08 | 2.10 | 13 | 0.61 | Moderate |
| | GorB-Tr | 4.94 | 2.01 | 25 | 0.59 | Moderate |
| May 2024 | Gor-C | 3.02 | 1.73 | 10 | 0.54 | Poor |
| | GorB-Tr | 2.78 | 2.54 | 19 | 0.75 | Good |
| November 2024 | Gor-C | 3.25 | 2.53 | 16 | 0.67 | Moderate |
| | GorB-Tr | 2.92 | 1.87 | 17 | 0.65 | Moderate |
| June 2025 | Gor-C | 3.33 | 2.35 | 14 | 0.63 | Moderate |
| | GorB-Tr | 3.00 | 1.91 | 11 | 0.57 | Moderate |

*Tab. 2. Community parameters and AMBI/M-AMBI values in the Sacca di Goro (Gor-C. Control; GorS-Tr: Seganda; GorB: Bassunsin transplant sites)*

The Seganda transplant site (GorS-Tr) was abandoned starting from 2024, as no evidence remained of the transplanted shoots. At the Bassunsin transplant site (GorB-Tr), the ecological quality, assessed through M-AMBI and BITS indices, was generally higher than that observed at the control site, and consistently lower percentages of opportunistic taxa were recorded. The temporal trend of the M-AMBI index is represented in Fig. 3, where, for each sampling date, the M-AMBI values found at the control site and at the transplant sites are compared.

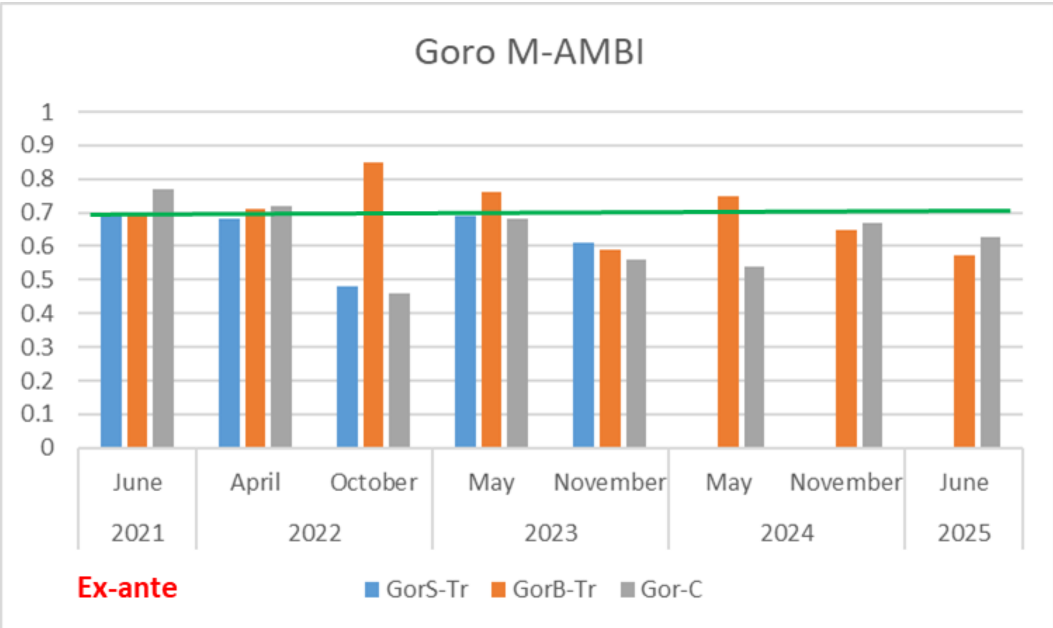


*Fig. 3. M-AMBI values at Goro (Gor-C. Control; GorS-Tr and GorB-Tr: Seganda and Bassunsin transplant sites). The green bar is the threshold between Moderate and Good ecological status*

**Table 3** shows the percentage of invertebrates belonging to the five sensitivity-tolerance groups: EG-I sensitive, EG-II indifferent, EG-III tolerant, EG-IV and EG-V opportunistic of second and first order respectively; the higher the percentage of EG-I (or the lower the percentage of EG-IV and EG-V), the better the ecological quality.

| *Date* | *Stations* | Ecological Groups | | | | |
|---|---|---|---|---|---|---|
| | | *I(%)* | *II(%)* | *III(%)* | *IV(%)* | *V(%)* |
| June 2021 | Gor-C | 4.34 | 1.19 | 92.71 | 0.39 | 1.38 |
| | GorS | 0.23 | 1.41 | 97.66 | 0.70 | 0 |
| April 2022 | Gor-C | 1.29 | 0.39 | 87.61 | 9.68 | 1.03 |
| | GorS-Tr | 1.45 | 0 | 79.42 | 14.78 | 4.35 |
| | GorB-Tr | 3.18 | 1.96 | 93.39 | 1.47 | 0.00 |
| October 2022 | Gor-C | 0 | 0.90 | 14.60 | 84.30 | 0.10 |
| | GorS-Tr | 0 | 0 | 91.70 | 8.00 | 0.40 |
| | GorB-Tr | 4.60 | 16.20 | 74.90 | 4.00 | 0.20 |
| May 2023 | Gor-C | 29.80 | 13.10 | 16.20 | 4.00 | 36.90 |
| | GorS-Tr | 0.80 | 0.30 | 61.50 | 37.00 | 0.40 |
| | GorB-Tr | 5.60 | 79.40 | 9.10 | 5.90 | 0.10 |
| November 2023 | Gor-C | 0 | 9.50 | 84.10 | 4.80 | 1.60 |
| | GorS-Tr | 0 | 0.0 | 97.0 | 0.40 | 2.60 |
| | GorB-Tr | 4.0 | 2.90 | 21.70 | 2.30 | 69.0 |
| May 2024 | Gor-C | 0.20 | 2.29 | 95.52 | 0 | 1.99 |
| | GorB-Tr | 6.76 | 50.37 | 16.47 | 3.66 | 22.74 |
| November 2024 | Gor-C | 2.68 | 1.39 | 79.78 | 9.05 | 7.11 |
| | GorB-Tr | 0.12 | 9.09 | 88.17 | 0.99 | 1.63 |
| June 2025 | Gor-C | 2.08 | 0.28 | 82.41 | 3.88 | 11.36 |
| | GorB-Tr | 3.68 | 0.50 | 87.86 | 7.78 | 0.18 |

*Tab. 3. Sensitivity-tolerance distribution of macrobenthos in the Sacca di Goro (Gor-C. Control; GorS-Tr: Seganda; GorB: Bassunsin transplant sites).*

The majority of the taxa present at the two transplant sites belongs to the “tolerant” group (EG-III), while at the control site to the opportunist group (EG-IV). At BAS, however, are present also sensitive (EG-I) and indifferent (EG-II) species. It is also evident how, in the transplant sites, the benthic community is better structured, in terms of ecological groups, than in the control site.

The application of the BITS index (**Table 4**) gave a higher value at the Bassunsin transplant site, where the ecological status was often Good (**Fig.4**). Both transplant sites exhibited, however, a better ecological status than the control site. It is well known that, in lagoon environments, the Ecological Status (ES) obtained using the BITS index is typically slightly higher than that derived from M-AMBI. This is because the BITS index was specifically developed for lagoonal communities, where the proportion of opportunistic taxa is naturally higher than in marine environments.

| *Date* | *Stations* | *EQR BITS* | *Status* |
|---|---|---|---|
| June 2021 | Gor-C | 0.44 | Moderate |
| | GorS | 0.52 | Moderate |
| April 2022 | Gor-C | 0.22 | Poor |
| | GorS-Tr | 0.50 | Moderate |
| | GorB-Tr | 0.80 | High |
| October 2022 | Gor-C | 0.25 | Poor |
| | GorS-Tr | 0.52 | Moderate |
| | GorB-Tr | 0.92 | High |
| May 2023 | Gor-C | 0.35 | Moderate |
| | GorS-Tr | 0.52 | Moderate |
| | GorB-Tr | 0.69 | Good |
| November 2023 | Gor-C | 0.40 | Moderate |
| | GorS-Tr | 0.54 | Moderate |
| | GorB-Tr | 0.89 | High |
| May 2024 | Gor-C | 0.80 | Good |
| | GorB-Tr | 0.51 | Moderate |
| November 2024 | Gor-C | 0.67 | Moderate |
| | GorB-Tr | 0.73 | Good |
| June 2025 | Gor-C | 0.57 | Moderate |
| | GorB-Tr | 0.77 | Good |

*Tab. 4. BITS EQR values in the Sacca di Goro (Gor-C. Control; GorS-Tr: Seganda; GorB: Bassunsin transplant sites).*

The ordination analysis using nMDS (**Fig. 5**) confirms the above, showing that the Bassunsin points (green triangles) from the transplant site (outlined with a solid red line) segregate separately from the controls (blue triangles), and also from Seganda transplant site (solid blu line).

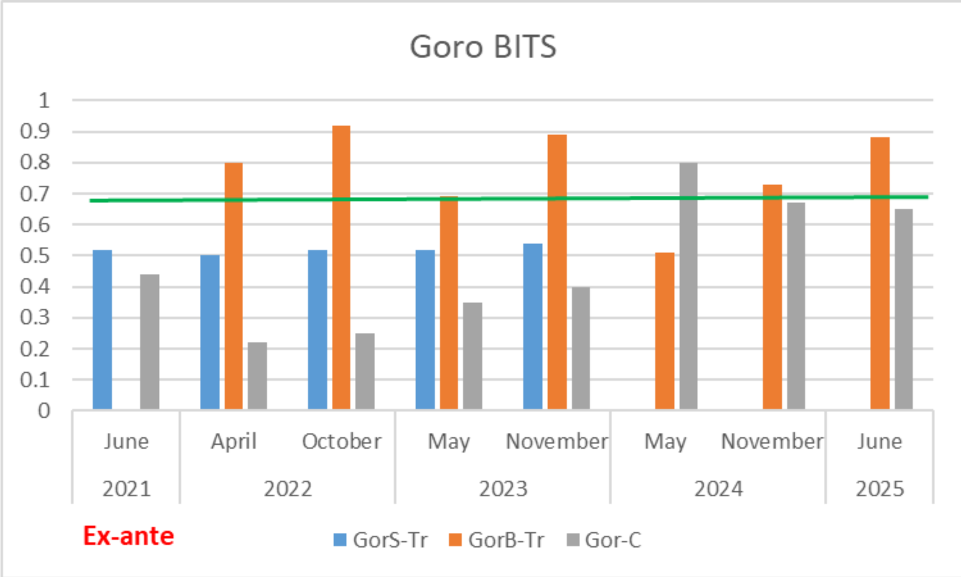



*Fig. 4. BITS values at Goro (Gor-C. Control; GorS-Tr and GorB-Tr: Seganda and Bassunsin transplant sites). The green bar is the threshold between Moderate and Good ecological status*

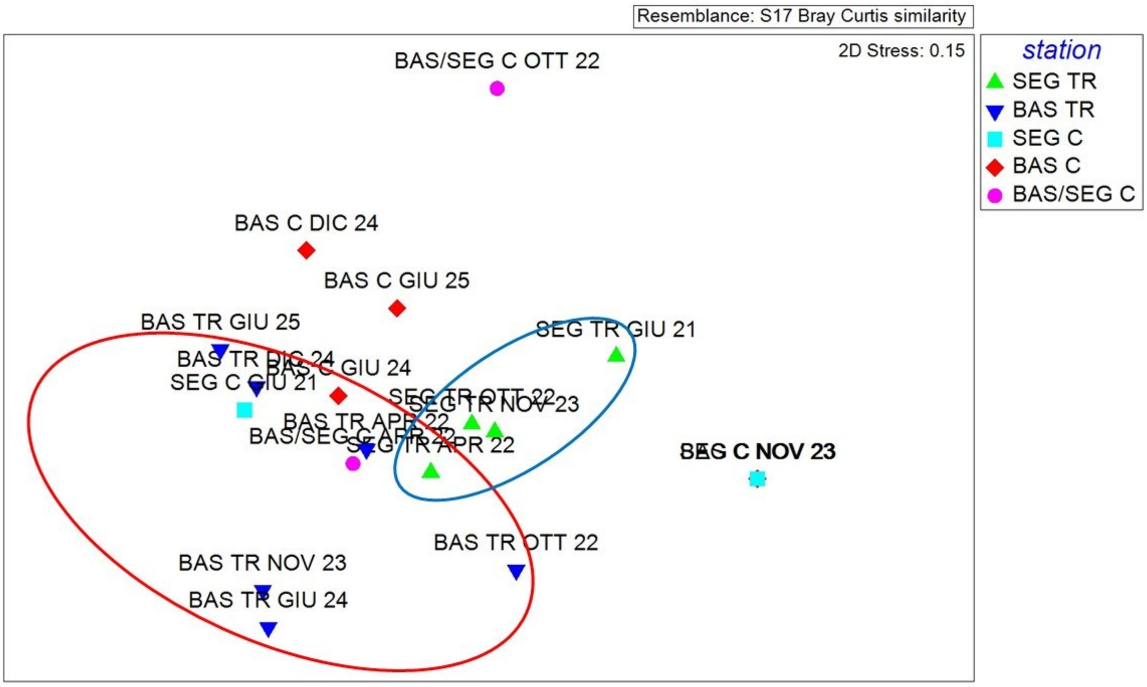



*Fig. 5. nMDS at Goro (Seg/Bas-C. Control; Seg-Tr and Bas-Tr: Seganda and Bassunsin transplant sites).*

**Valle Fattibello lagoon**

Overall, 33 macrobenthic taxa were found in the Valle Fattibello. **Table 5** shows the faunal list of the macrobenthic taxa collected during the monitoring campaign.

| | |
|---|---|
| *Alitta succinea* | *Gammarus aequicauda* |
| *Capitella capitata* | *Gammarus insensibilis* |
| *Fabricia sp.* | *Grandidierella japonica* |
| *Fabricia stellaris* | *Grandidierella sp* |
| *Hediste diversicolor* | *Idotea balthica* |
| *Heteromastus filiformis* | *Melita palmata* |
| *Oligochaeta* | *Monocorophium acherusicum* |
| *Polydora cornuta* | *Monocorophium insidiosum* |
| *Prionospio cirrifera* | *Arcuatula senhousia* |
| *Spio filicornis* | *Bittium reticulatum* |
| *Streblospio eridani* | *Donax trunculus* |
| *Streblospio shrubsolii* | *Hydrobia acuta* |
| *Amphibalanus* | *Hydrobia sp* |
| *Carcinus maenas* | *Hydrobia ventrosa* |
| *Chironomus salinarius* | *Ruditapes philippinarum* |
| *Corophium orientale* | *Actinia spp* |
| *Cyathura carinata* | |

*Tab. 5. Taxonomic list of the macrobenthic taxa identified in the Valle Fattibello*

In **Table 6** the values of community descriptors (diversity and species richness), together with AMBI/M-AMBI values and ES are reported. In the Table also the reference parameters are shown (in grey).

| *Date* | *Stations* | *AMBI* | *H'* | *S* | *M-AMBI* | *Status* |
|---|---|---|---|---|---|---|
| | Bad | 6 | 0 | 0 | 0 | Bad |
| | High | 1.85 | 3.3 | 25 | 1 | High |
| June 2021 | Fat-C | 2.78 | 2.19 | 30 | 0.84 | Good |
| | Fat | 3.33 | 1.55 | 13 | 0.54 | Poor |
| April 2022 | Fat-C | 2.78 | 2.19 | 29 | 0.84 | Good |
| | Fat-Tr | 2.89 | 2.93 | 24 | 0.86 | Good |
| October 2022 | Fat-C | 3.64 | 2.29 | 17 | 0.65 | Moderate |
| | Fat-Tr | 3.64 | 2.12 | 16 | 0.62 | Moderate |
| May 2023 | Fat-C | 3.35 | 2.68 | 18 | 0.70 | Moderate |
| | Fat-Tr | 3.05 | 2.66 | 15 | 0.69 | Moderate |
| November 2023 | Fat-C | 3.05 | 3.04 | 13 | 0.71 | Good |
| | Fat-Tr | 2.92 | 2.21 | 18 | 0.71 | Good |

| | | | | | | |
|---|---|---|---|---|---|---|
| May 2024 | Fat-C | 3.61 | 2.82 | 12 | 0.63 | Moderate |
| | Fat-Tr | 3.27 | 2.16 | 19 | 0.69 | Moderate |
| November 2024 | Fat-C | 3.21 | 2.26 | 16 | 0.67 | Moderate |
| | Fat-Tr | 3.04 | 1.96 | 12 | 0.59 | Moderate |
| June 2025 | Fat-C | 3.32 | 1.97 | 9 | 0.54 | Poor |
| | Fat-Tr | 3.00 | 1.90 | 17 | 0.66 | Moderate |

*Tab. 6. Community parameters and AMBI/M-AMBI values at Fattibello (Fat-C: control; Fat-Tr: transplant site)*

The temporal trend of the M-AMBI index is represented in **Fig. 6**, where, for each sampling date, the M-AMBI values found at the control site and at the transplant site are compared.

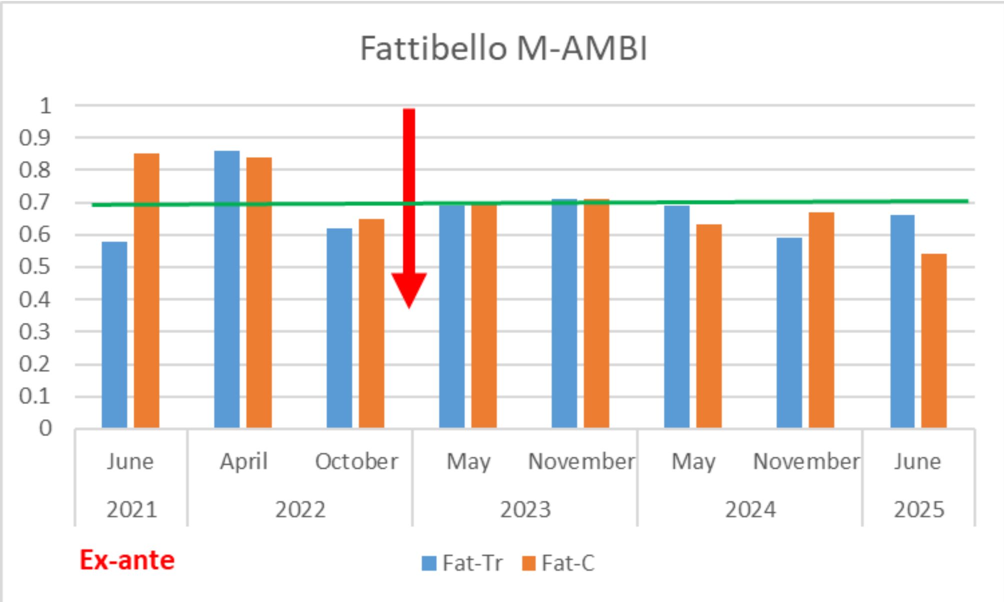



*Fig. 6. Ecological quality through M-AMBI at Fattibello (Fat-C: control; Fat-Tr: transplant site). The green bar is the threshold between Moderate and Good ecological status; the red arrow indicates the change in the transplant site*

**Table 7** shows the percentage of invertebrates belonging to the five sensitivity-tolerance groups: EG-I sensitive, EG-II indifferent, EG-III tolerant, EG-IV and EG-V opportunistic of second and first order respectively; the higher the percentage of EG-I (or the lower the percentage of EG-IV and EG-V), the better the ecological quality.

| *Date* | *Stations* | Ecological Groups | | | | |
|---|---|---|---|---|---|---|
| | | *I(%)* | *II(%)* | *III(%)* | *IV(%)* | *V(%)* |
| June 2021 | Fat-C | 9.00 | 3.20 | 82.90 | 3.20 | 1.70 |
| | Fat | 0.10 | 0.10 | 87.80 | 1.50 | 10.50 |
| April | Fat-C | 9,00 | 3,20 | 82,90 | 3,20 | 1,70 |

| | | | | | | |
|---|---|---|---|---|---|---|
| 2022 | Fat-Tr | 5,54 | 3,97 | 82,84 | 7,66 | 0 |
| October | Fat-C | 0 | 7,7 | 43,8 | 46,5 | 1,9 |
| 2022 | Fat-Tr | 0 | 7,8 | 42,7 | 48,5 | 0,9 |
| May | Fat-C | 5.7 | 1.4 | 58.7 | 32.4 | 1.7 |
| 2023 | Fat-Tr | 0.3 | 21.2 | 53.8 | 24.3 | 0.3 |
| November | Fat-C | 2.4 | 15.3 | 69.4 | 2.4 | 10.6 |
| 2023 | Fat-Tr | 6 | 0 | 90.7 | 0.2 | 3.2 |
| May | Fat-C | 1.96 | 0.0 | 75.63 | 0.11 | 22.31 |
| 2024 | Fat-Tr | 1.47 | 0.10 | 87.87 | 0.10 | 10.46 |
| November | Fat-C | 0.99 | 0.60 | 89.99 | 0.10 | 8.33 |
| 2024 | Fat-Tr | 1.59 | 0 | 95.43 | 0.05 | 2.94 |
| June | Fat-C | 0.19 | 0.0 | 88.89 | 0.00 | 10.92 |
| 2025 | Fat-Tr | 0.26 | 0.0 | 99.37 | 0.28 | 0.09 |

*Tab. 7. Sensitivity-tolerance distribution of macrobenthos at Fattibello (Fat-C: control; Fat-Tr: transplant site)*

The majority of taxa present at the two sites belongs to the “tolerant” (EG-III), however there are very strong fluctuations in the composition of the community between the various periods and the 2 sites.

The application of the BITS index (**Table 8**) generally gave the similar ES values at both sites.

| *Date* | *Stations* | *EQR BITS* | *Status* |
|---|---|---|---|
| June | Fat-C | 0.85 | Good |
| 2021 | Fat | 0.64 | Moderate |
| April | Fat-C | 0.70 | Good |
| 2022 | Fat-Tr | 0.77 | Good |
| October | Fat-C | 0.63 | Moderate |
| 2022 | Fat-Tr | 0.60 | Moderate |
| May | Fat-C | 0.55 | Moderate |
| 2023 | Fat-Tr | 0.57 | Moderate |
| November | Fat-C | 0.79 | Good |
| 2023 | Fat-Tr | 0.81 | Good |
| May | Fat-C | 0.40 | Poor |
| 2024 | Fat-Tr | 0.55 | Moderate |
| November | Fat-C | 0.69 | Good |
| 2024 | Fat-Tr | 0.84 | Good |
| June | Fat-C | 0.52 | Moderate |
| 2025 | Fat-Tr | 0.69 | Good |

*Tab. 8. BITS EQR values at Fattibello (Fat-C: control; Fat-Tr: transplant site)*

The following Fig. 7 shows the temporal trane of BITS values at Fattibello.

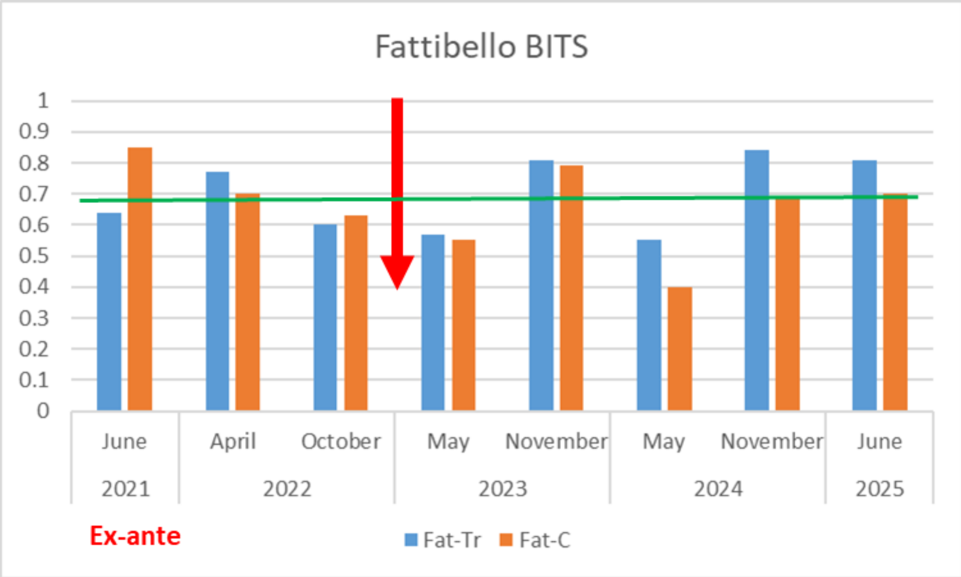



*Fig. 7. Ecological quality through BITS at Fattibello (Fat-C: control; Fat-Tr: transplant site). The green bar is the threshold between Moderate and Good ecological status; the red arrow indicates the change in the transplant site*

Data from May 2023 onwards are always referred to the new transplant site (see **Fig. 8**).


*Fig. 8. New transplant site at Fattibello*

The ordination analysis using nMDS (**Fig. 9**) however shows that the station points are fairly separated, with a certain degree of segregation of the points related to the transplant site (green triangles), especially those related to winter and summer 2024 and 2025, from the others.

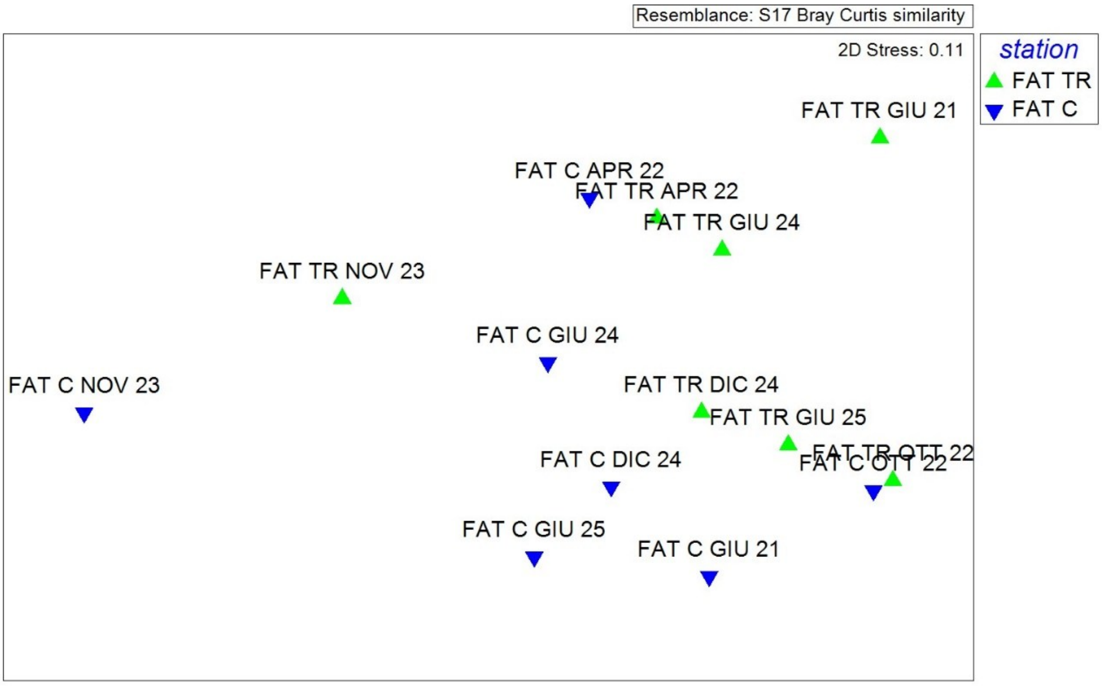



*Figure 9. nMDS at Fattibello Lagoon (Fat-C: control site; Fat-Tr: transplant site).*

## Conclusions

### Ecological Quality Assessment Using M-AMBI and BITS Indices

Although the aquatic angiosperm transplantation efforts carried out in the Goro and Fattibello lagoons achieved only limited success in terms of plant establishment and spatial coverage, the monitoring data collected indicate some positive ecological responses in the benthic compartment. In particular, since November 2024 at Goro and May 2024 at Fattibello (with the exception of November 2024), the ecological quality, as assessed through the application of macrobenthic indices, has been found to be slightly higher in the transplant sites compared to their respective control sites.

This observation, while modest in magnitude, suggests that even the small and fragmented patches of aquatic angiosperms that have managed to establish may have contributed to improving local environmental conditions and influencing the structure of the macrobenthic community. The presence of angiosperms, even in limited extent, can locally enhance sediment stability, increase oxygenation near the sediment–water interface, and provide structural complexity that favors the settlement of more diverse and less opportunistic benthic assemblages.

Analysis of the macrobenthic community composition in both lagoons confirms this tendency. Although the proportion of sensitive taxa (belonging to ecological groups EG-I and EG-II) remains low in absolute terms, reflecting the overall degraded or transitional status of the sites, the relative abundance of opportunistic taxa (EG-IV and EG-V) is consistently higher in the control areas than in the transplant sites. This pattern indicates that the presence of angiosperm patches may already be exerting a stabilizing effect on the benthic environment, reducing the dominance of opportunistic species typically associated with organically enriched or disturbed conditions.

From an ecological perspective, these results are encouraging. They suggest that even limited angiosperm establishment can act as an incipient driver of ecological recovery, promoting a gradual shift in macrobenthic community composition toward assemblages characteristic of better ecological quality states. Although the overall magnitude of change is still small and further monitoring will be necessary to confirm long-term trends, the observed differences between transplant and control sites highlight the potential functional role of these emerging angiosperm patches in supporting benthic biodiversity and ecosystem resilience.

## Fish fauna and HBFI determination

***Methods***

***The HFBI (Habitat Fish Bio Indicator) index***

The HFBI is an empirically derived multi-metric index, composed of six metrics expressed as ecological quality reports. The index is calculated by combining various ecological descriptors including species richness and biomass, but also characteristics related to the belonging of each species to the different functional groups. They contribute to the calculation of the metrics that describe the characteristics of the functional groups, exclusively species belonging to the following ecological guilds: estuarine residents (ES), diadromes (Di) and marine migratory (MM). These groups were included due to their high susceptibility to environmental degradation, being highly dependent on the integrity of the habitats for the purposes of reproduction, nourishment and growth.

The fishing tool we adopted has such a selectivity as to allow a representative sampling of the fish communities present in the sampled area and associated with the particular type of habitat to be monitored. The mesh size (internode equal to 2 mm) of the tool allow to capture even small species such as *Aphanius fasciatus* and *Syngnathus abaster,* often associated with environments in good health. Furthermore, since in the lagoon environments the distribution of fish species is strongly influenced by some environmental variables, such as water temperature, salinity, oxygen dissolved as well as vegetation cover, and has a strong seasonal variability, the index was designed in such a way as to be able to evaluate the structure of the fishing communities according to the type of water body, seasonality (spring and autumn) and habitats (vegetated or non-vegetated environment).

Sampling was carried out in Goro and Fattibello at July and November 2022, May and November 2023, May and November 2024, and finally June 2025.

**Results**

**Table 9** shows the taxonomic list of fish species captured in the Sacca di Goro, while **Table 10** is referred to Fattibello fish community. At Goro the fish community was always richer (both in species number and abundance) and more diversified respect to Fattibello, probably reflecting the different level of confinement respect to the sea of the 2 lagoons. In general, in Goro the fish community is dominated in weight and abundance by mullets, while in Fattibello by sandsmelts and gobies.

| | |
|---|---|
| *Anguilla anguilla* | *Pomatoschistus canestrini* |
| *Atherina boyeri* | *Zosterisessor ophiocephalus* |
| *Sardina pilchardus* | *Dicentrarchus labrax* |
| *Alosa fallax* | *Chelon auratus* |
| *Sprattus sprattus* | *Liza ramada* |
| *Aphanius fasciatus* | *Solea solea* |
| *Engraulis encrasicolus* | *Syngnathus abaster* |
| *Knipowitschia panizzae* | *Mullus barbatus* |
| *Gobius niger* | *Chelon saliens* |

*Table 9. Fish species at Goro*

| | |
|---|---|
| *Atherina boyeri* | *Knipowitschia panizzae* |
| *Sardina pilchardus* | *Gobius niger* |
| *Sprattus sprattus* | *Pomatoschistus canestrini* |
| *Aphanius fasciatus* | *Zosterisessor ophiocephalus* |
| *Engraulis encrasicolus* | *Solea solea* |

*Table 10. Fish species at Fattibello*

The application of the HFBI index is shown in **Tables 11 and 12.** At both lagoons, at the transplant sites the value of the index was slightly higher respect to the control site.

| *Date* | *Stations* | *HBFI* | *Status* |
|---|---|---|---|
| July 2022 | Gor-C | 0.85 | Good |
| | GorB-Tr | 1.0 | High |
| November 2022 | Gor-C | 0.8 | Good |
| | GorB-Tr | 1.0 | High |
| May 2023 | Gor-C | 0.23 | Poor |
| | GorB-Tr | 0.28 | Poor |
| November 2023 | Gor-C | 0.85 | Good |
| | GorB-Tr | 0.99 | High |
| May 2024 | Gor-C | 0.79 | Good |
| | GorB-Tr | 0.80 | Good |
| November 2024 | Gor-C | 0.51 | Moderate |
| | GorB-Tr | 0.68 | Good |
| June 2025 | Gor-C | 0.51 | Moderate |
| | GorB-Tr | 0.64 | Good |

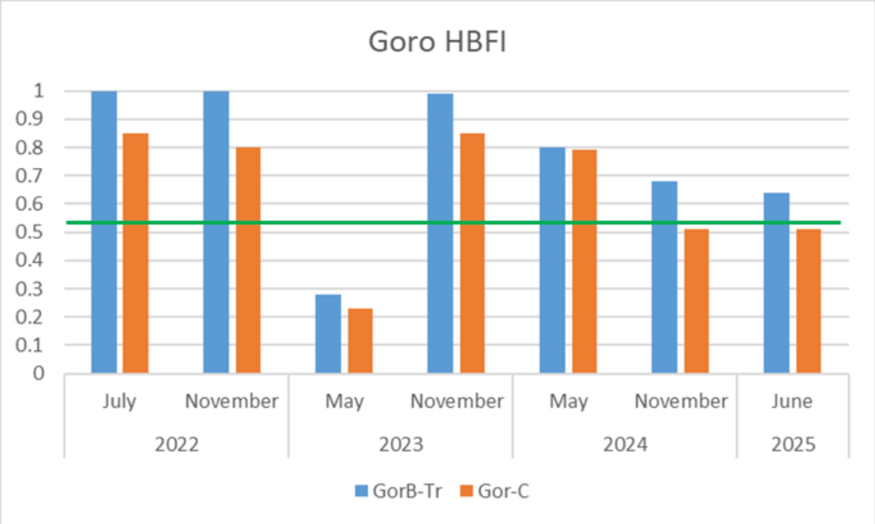
*Table 11. HBFI at Goro*

| *Date* | *Stations* | *HBFI* | *Status* |
|---|---|---|---|
| June 2022 | Fat-C | 0.38 | Moderate |
| | Fat-Tr | 0.30 | Poor |
| November 2022 | Fat-C | 0.49 | Moderate |
| | Fat-Tr | 0.40 | Moderate |
| May 2023 | Fat-C | 0.15 | Poor |
| | Fat-Tr | 0.14 | Poor |
| November 2023 | Fat-C | 0.25 | Poor |
| | Fat-Tr | 0.36 | Moderate |
| May 2024 | Fat-C | 0.25 | Poor |
| | Fat-Tr | 0.87 | High |
| November 2024 | Fat-C | 0.33 | Moderate |
| | Fat-Tr | 0.33 | Moderate |
| June 2025 | Fat-C | 0.45 | Moderate |
| | Fat-Tr | 0.65 | Good |

*Table 12. HBFI at Fattibello*

**Fig. 10** shows the temporal trend of HBFI at Goro and Fattibello. The application of the HBFI (Habitat-Based Fish Index) resulted in Ecological Status (ES) values that were lower than expected. The HBFI is a multimetric index specifically designed to assess the ecological quality of transitional and coastal waters through the analysis of fish assemblages. It integrates several ecological components, including species richness, abundance, trophic structure, and the relative contribution of sensitive and opportunistic taxa, with results expressed on a five-class scale of ecological status.

In our case, the relatively low ES values, especially at Fattibello, obtained from HBFI appear to be influenced primarily by methodological constraints rather than by actual ecological factors. Specifically, the sampling followed the protocol developed by ISPRA (2017), which is the standardized method currently adopted at the Italian national level. However, this approach may not be fully appropriate for highly dynamic lagoon systems, particularly in areas located near the sea inlets. In such environments, fish assemblages are characterized by high mobility, rapid turnover, and pronounced seasonal and tidal migrations.



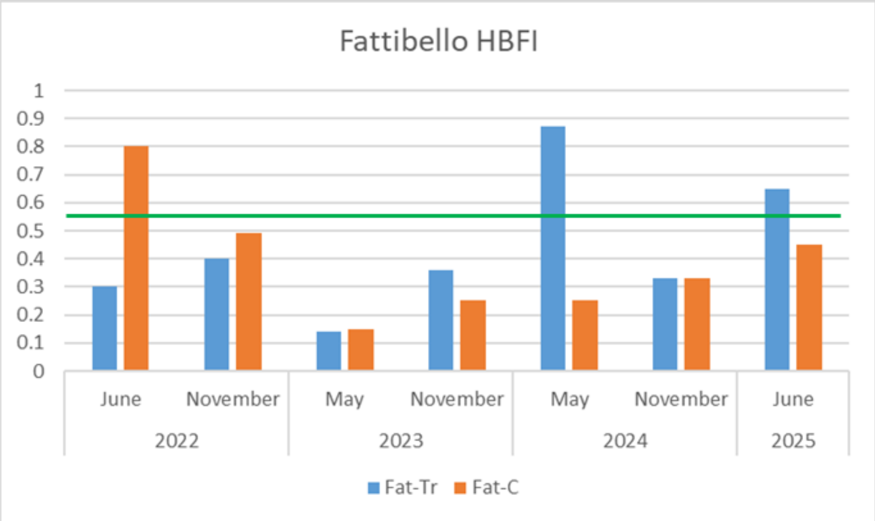




*Figure 10. HBFI values at Goro and Fattibello Lagoons. The green bar is the threshold between Moderate and Good ecological status*

Under these conditions, the standard gear employed, a 10-meter-long seine net, may inadequately represent the true composition and abundance of the fish community, potentially leading to an underestimation of the Ecological Status. The limited sampling coverage and short capture radius of the net are likely insufficient to effectively sample transient or fast-moving species that dominate these transitional habitats.

The better ecological quality observed at Goro may also be related to the fact that Goro is an open-type lagoon, whereas Fattibello is a choked-type system. The greater marine influence at the Bassunsin site in Goro, located near the lagoon inlet, likely allows for higher fish fauna richness, independently of the presence of the limited patches of *Nanozostera*

**Figure 11** shows some phases of the fishing activity.

*Fig. 11. Fishing with the shore seine*

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