Continuous Nutrient automated monitoring on the Mediterranean Sea using in situ flow analyser

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Abstract- **The development of field instruments for in situ chemical analysis in seawater is essential to better understand the physico-chemical processes occurring in the ocean as well as the supervision of the anthropogenic impact on the marine ecosystem in coastal and open ocean. Temporal resolution sampling for the analysis of chemical parameters in aquatic systems is usually low (weekly or monthly), expensive and time consuming. Some important details of transient phenomenon such as night and day cycling, strong tidal currents, episodic weather events and algal blooms can be missed. One of the advantages of in situ chemical measurements is the achievement of long term data sets to better characterize the natural variability of these elements. The use of in situ chemical analyzers also minimizes the probability of chemical changes in sample, due to reactions or contamination that may occur during transfer and storage of collected samples. There is actually the need of rugged, portable, fully automated instrumentation that can operate in situ, in order to avoid collection, transport and storage of samples for laboratory analysis; the availability of such instrumentation has proven to be a difficult challenge. Recently the WIZ multiparametric in-situ probe was developed by SYSTEA, to measure sequentially up to four chemical compounds in surface and sea water. It has been tested during two weeks in the Mediterranean sea near the French coastal station of Observatoire Océanologique de Banyuls (OOB) from University Pierre et Marie Curie (UPMC, Paris VI) in Banyuls sur Mer, well known as "Laboratoire ARAGO" since its creation two centuries ago. The probe has been deployed at 2m depth on the coastal buoy developed by OOB, to be installed first in a point which is surveyed weekly since 1997 (SOLA station, 42°29'300 N – 03°08'700 E) with manual sampling and laboratory analysis. This area is particularly interesting due to the oligotrophic conditions which allowed to estimate the probe sensitivity and selectivity. In the near future, the Banyuls Observatory will deploy this automated buoy at 20 miles from the coast (MOLA station), in order to make a high frequency and long term survey of the area using several sensors. This operation will be done according to the French National program for Mediterranean Sea survey network MOOSE (Mediterranean Ocean Observation multi-Sites on Environment).**

Keywords: Continuous nutrients monitoring, autonomous observatory systems, in-situ multiparametric analyzers WIZ probe.

I. INTRODUCTION

Continuous monitoring of a highly dynamic system such as coastal waters requires frequent sampling at a precise place in order to catch short-term events which might have a strong impact on the coastal ecosystem, such as exceptional phytoplankton blooms or change caused by storms. The lack of monitoring systems that provide continuous observations of the marine environment, especially concerning chemical species like nutrients in the coastal areas and shelf seas of Europe, is a serious hindrance. Instruments for in-situ dissolved nutrient analyses are needed by oceanographers. In-situ measurement techniques have many advantages, like avoiding the use of storage bottles to be shipped to land-based laboratories, reducing the risk of sample contamination. In any case, additional and collateral sampling can be performed when special phenomena are identified.

Potential applications of submersible analysers include pollution control, effluent monitoring and nutrient loads studies. Nutrients commonly measured during field studies include nitrate, nitrite, ammonia, phosphate, silicate, dissolved organic nitrogen and phosphorus species.

Most in situ nutrient analyzers use wet chemical techniques based on laboratory methods developed during the last century. Wet chemical analyzers such as ALCHIMIST [4], ANAIS, [1], SubChemPak [2], Envirotech NAS-2, NAS-3 and Ecolab, SYSTEA NPA and DPA [13] and CHEMINI [3] require the addition of chemical reagents to the sample, to determine one or multiple nutrients (nitrate, nitrite, phosphate, ammonia, iron) Such analyzers exhibit good accuracy due to the use of onboard standards, which can be analyzed separately from the samples to provide a reference measurement. Independent measures of the sample, standards and reagents may be used to correct drifts and degradation in the optics or in the reagents.

So far, however, automated monitoring of marine water is not well developed due to the difficult conditions in the sea during storms or strong tidal currents. Moored automated systems have been deployed offshore; paradoxically, although their maintenance is easier in coastal waters than in offshore waters, only a few automated buoys have been moored in the coastal environment for long-term measurements because of sensors bio-fouling [5].

The laboratory ARAGO was settled on the "Côte Vermeille" since 1882 and it is one of the most ancient marine research stations of France and Europe. Since its creation this laboratory continuously developed field observation in combination with its own research activities. In 1997, the "Service of Observation" of the Laboratory Arago (SOLA) was created; it is also member of the National Service of Observation in LITTORAL Environment (SOMLIT), to start a long-term physico-chemical measurement protocol common to several French coast stations, in agreement with the objectives fixed by the project MOOSE (Mediterranean Ocean Observation multi-sites on Environment) supported by INSU (Institut National des Sciences de l'Univers) within the framework of a Mediterranean Sea construction site.

Nutrient probes deployment history on autonomous buoys for coastal monitoring

During the last ten years, several implementations of SYSTEA nutrient analysers have been done in collaboration with the French institute IFREMER in marine autonomous system from the French automated network ROSLIT and using MAREL (Mesures Automatisées en Réseau de l'Environnement Littoral) buoy technology [6] (http://www.ifremer.fr/dtmsi/programmes/marel/marel.htm). A test of implementation has been done at IFREMER with a first generation NPA (Nutrients Probe Analyzer) and it was later installed in the harbour of Boulogne-sur-Mer in the MAREL Carnot station starting from November 25th, 2004. This station was measuring every 20 minutes nutrients, salinity, seawater and air temperature, fluorescence, turbidity, dissolved oxygen, PAR, and wind speed and direction for five years now. This automated station is fixed on a dike (Figure 1) and it consists in a fifteen meters tube containing a floating basement, supporting a tidal range is 10 meters where all the different sensors and a data-logging system are mounted (Figure 2).

Fig. 2 MAREL CARNOT autonomous station

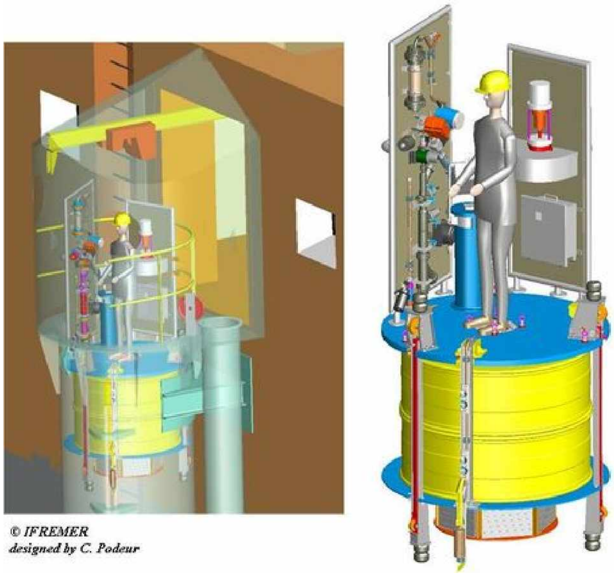

Fig. 2 MAREL CARNOT autonomous station drawing with NPA implemented (blue cylinder)

A six month validation test has been done measuring nitrate, nitrite, phosphate and silicate concentrations every 12 hours. Change of reagents and calibration standards were done every month. Results have shown a very good correlation between nutrients and other physical parameters like salinity and fluorescence, which is an indicator for chlorophyll abundances (Figure 3). Two analyzers were used making a rotation every month during maintenance operations, in order to ensure a complete clean up and recalibration. In Figure 3 is shown the good correlation between the increase in nutrients concentration and the reduction of salinity, due to flow of fresh water entering the Boulogne harbour. In the same figure it can be observed the perfect correlation between phytoplankton bloom and decrease in concentration of nutrients consumed during the photosynthesis phenomenon. This experience allowed studying the influence of continental water contribution and especially of the nutrient load coming from Liane river catchment basin affecting the creation of small algal bloom [7].

This long term deployment demonstrated to the scientific French community the feasibility and the interest to apply a nutrient analyzer to detect anthropic effects in a high frequency mode; weekly manual samplings, instead, do not permit to detect these kinds of phenomena. Another important result was the proven reliability of the NPA probe during this long term deployment. However, additional efforts in system's miniaturisation and in minimizing power and reagents consumptions were necessary for long term deployments on coastal buoys, where power supply and maintenance access are more limited.

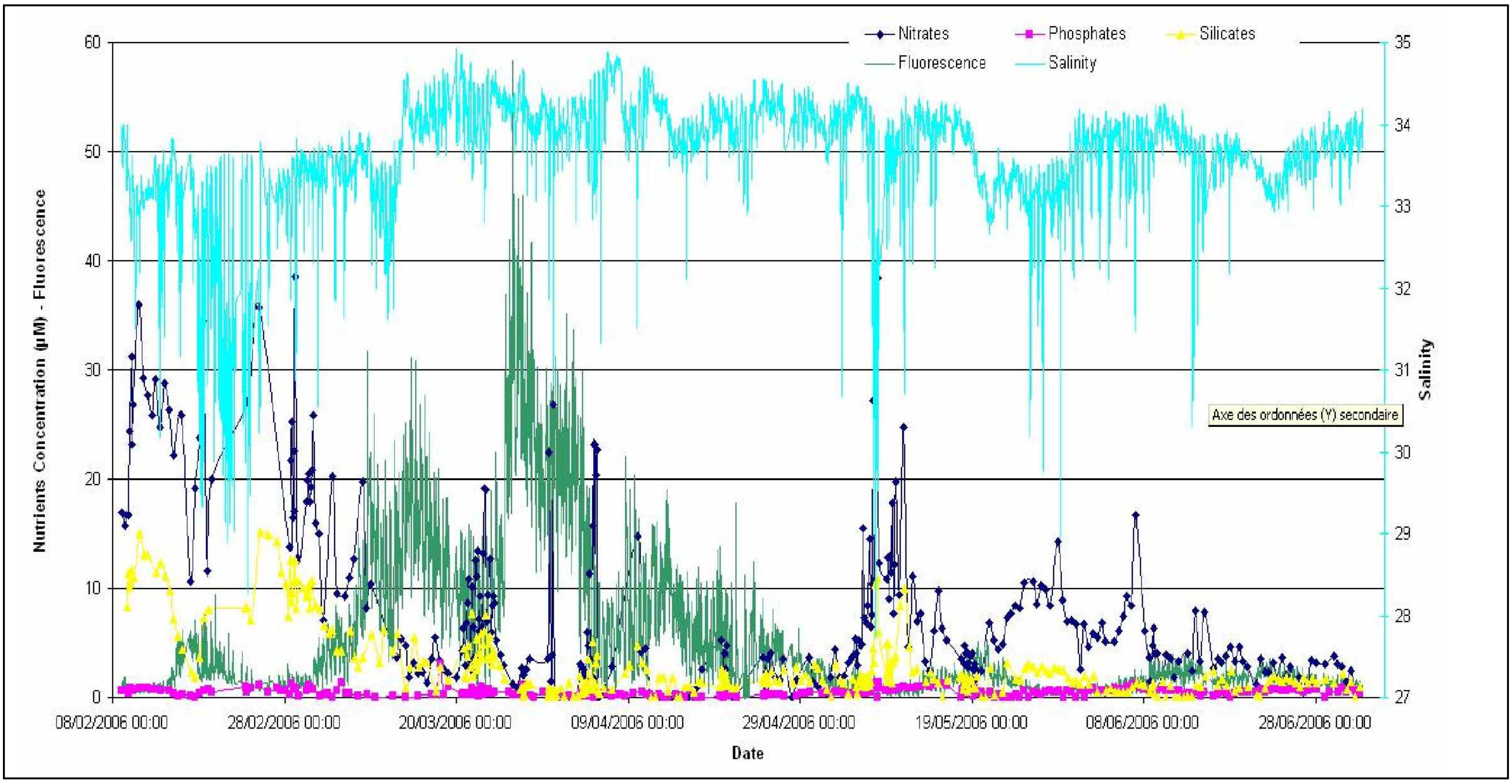

Fig. 3 Nutrient results obtained with NPA probe during 4.5 months period in Boulogne sur Mer

Needs for autonomous automated observatories

Solutions for autonomous automated observatory stations implementations should rely on multi-purpose, multi-scale intelligent, adaptive, automated, network enabled, inter-operable low cost, low maintenance technologies. Most criteria are mandatory in an operational oceanography framework but also for climate research and water quality survey on the long-term. Operational oceanography networks are collecting routinely in-situ and remote sensing hydrodynamic data, including sometimes multi-disciplinary (i.e. biology, optical) marine environment parameters. Remote sensing data offer regular depiction of the sea surface, while in-situ data provide full water column scarce spatial information. Marine observatories in the European countries are integrated within dedicated networks. Regional efforts on the integration of marine observations need to be consolidated through capacity building and technology transfer efforts.

The scientific French community is now getting organized at many levels by dedicated infrastructure developments and regular technology watch should become a standard activity. That's one of the goals of the MOOSE (Mediterranean Ocean Observation multi-Sites on Environment) French project for the Mediterranean Sea [8].

The ocean environmental is by definition hostile to sensors and there is space for improvements on many issues. Bio-fouling limit the period an instrument can be deployed in water but anti-fouling paints are usually not environment friendly. Except for cabled systems, usual platforms have limited lifetime, but new models of batteries (e.g. capacitor, fuel cells) will extend it significantly. Artificial intelligent sensors may optimize idle modes, trigger specific sampling patterns and report malfunctioning. Increased storage capacities and communications have decreased the recovery/redeployment rate of moorings. New developments on underwater positioning systems, acoustic, optical and magnetic communications and inductive battery recharging are expected in the near future. Nanotechnologies will also decrease the energy demand of miniaturized optical and geo-chemical sensors (i.e. micro arrays, cameras, flow-cytometry, green-house gas detection).

Cooperative behavior and fleet mission planning tools for autonomous platforms will greatly enhance the effectiveness of monitoring and require the definition of international standards to be inter-operable. Issues like sensor inter-calibration could jeopardize a whole experiment and require utmost attention. Data management may be centralized or distributed depending on the end-user requirements but has to cope with increased volume and diversity of data of different kind. Automatic sensor calibration, data quality check procedures and techniques with limited human intervention should be preferred. Grid computing, data mining techniques and new multi-dimensional visualization tools will offer new ways to looks at complex data sets and modeling products, which should be disseminated on the world-wide web using user-friendly tools.

More sensors are expected in the future for chemical parameters measurement and technologies should be flexible enough to accommodate future innovations. Due to these considerations and the above reported positive experience, the Observatoire Océanologique de Banyuls established a technical and scientific collaboration with SYSTEA about in situ analyzers. The use of new UV sensors (like ISUS or ProPS-UV) for nitrate measurement showed bad adaptation to oligotrophic conditions in the Mediterranean Sea. In-flow analysis remains the best way to detect such low concentrations using the well known analytical methods based on spectrophotometry and fluorimetry to detect various dissolved chemical species (and not only nitrates), allowing the use of onboard standards to recalibrate *in situ* the probe.

II. CONTEXT AND MATERIALS

The Observation Service network

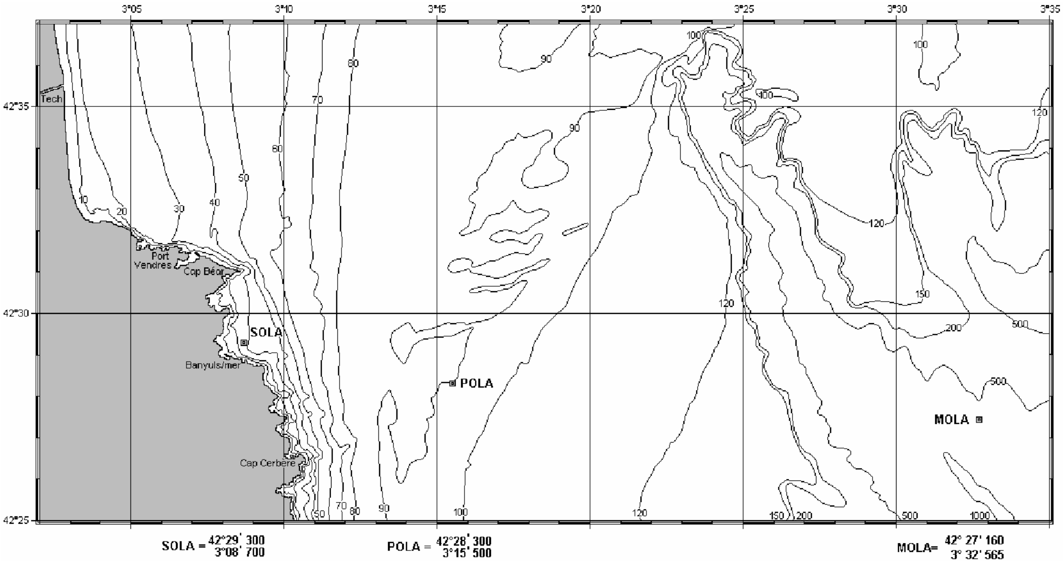

Fig. 4 Laboratoire ARAGO stations network

The Observation Service works on a network of three stations (Figure 4):

SOLA is the station realized for the SOMLIT network; it is placed in 0.5 nautical mile from the Observatory (42°29 ' 300 N - 03°08 ' 700 E, depth 27m)

POLA is placed in 5 nautical miles from the Observatory (42°28'300 N – 03°15'500 E, depth 95m). This station is located on the continental shelf and it allows observations on dense water formation and particulate transfer to the canyon.

MOLA is situated on the north side of the canyon Lacaze-Duthiers, at 19 nautical miles from the coast (42°27 ' 136 N - 03°32 ' 468 E, depth 600m). This station can be influenced both by waters from the Gulf of Lion or by those from the Mediterranean north current.

MOLA buoy description

The monthly sampling periodicity at the MOLA station is often disturbed due to bad weather conditions. Instrumented platforms are required for the real-time monitoring of environmental changes and to better assess biological processes. Owing to rapid turnover and growth rates, microbial systems (viruses, bacteria and phytoplankton) respond quickly (abundance, activity, diversity) to environmental changes and they are relevant indicators of modifications in the carbon sequestration. Incorporation of meteorological, hydrodynamic and biogeochemical measurements at time scales relevant for the processes related to the functioning of the microbial loop is a necessary and fundamental advance for the improvement and validation of coupled hydrodynamic-biogeochemical models. The MOLA station has to become soon an instrumented autonomous observatory for long term deployment and high frequency sampling with a real-time access to data. The platform is a MOBILIS buoy (diameter 3m, height 5.5m). A pyramidal structure has been developed and integrated to support all the external sensors (meteorological station and GPS), position light, power supply (solar panels and batteries), data-logger unit and remote communication device using WIFI technology. A SeaBird SBE16 CTD probe is placed below the flotation elements on the central mast for surface salinity, temperature, fluorescence and turbidity data measurement (Figure 5).

Fig. 5 MOLA automated buoy

To simplify the maintenance operations on the sensors, a second mooring line will be installed near the platform which will integrate a set of SeaBird MICROCAT CTD probes in fixed points at various depths or a profiling system like the Mac Lane MMP. This second mooring line will be implemented with an acoustic releaser for recovery and maintenance and with an acoustic modem for communication with the buoy (Figure 6).

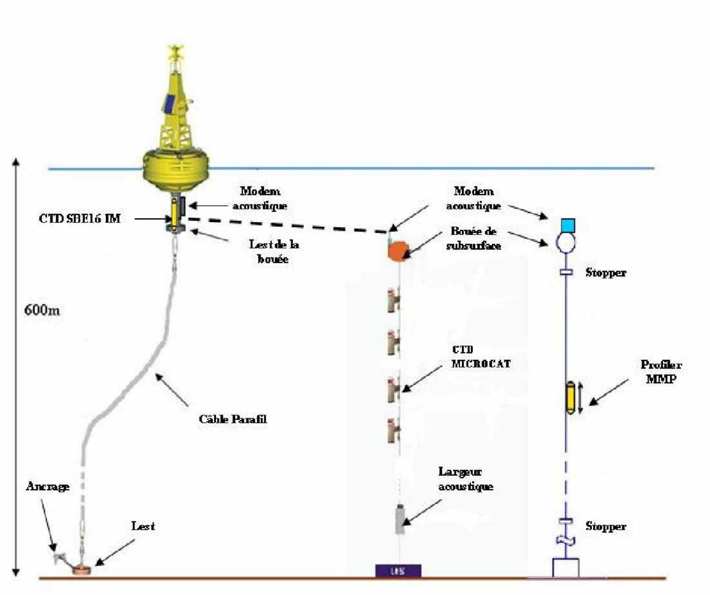

Fig. 6 MOLA buoy final design

WIZ probe deployment

The WIZ probe (Figure 7) is a real advanced analytical probe for "in-situ" application. It includes a 1.5 ml micro Loop Flow Reactor (LFR), enabling an extremely low consumption of reagents and standard, a multibeam optic fibers based colorimetric detector coupled with a microfluorimeter. An innovative "plug-in" and compact reagents container allows an immediate field reagents and calibration solutions changeover. The PVC single body is 140 mm diameter and 720 mm height, including the reagents container: the instrument's miniaturization has increased the ease of handling; the compact cylindrical design allows the deployment similar to conventional in-situ water quality monitoring probes.

Power consumption has been dramatically decreased (3W stand-by, 6 W analysis, max 1 A, 12 Vdc) compared to the previous NPA probe.

The two main analytical innovations are: the use of a fluorimetric method for ammonia detection [9], ensuring high selectivity and sensitivity minimizing matrix effects [10]; the use of UV photo reduction and subsequent determination of reaction products as nitrites is applied for nitrate detection [11, 12].

Fig. 7 WIZ probe tested in lab and the hand-held reagents container

Results are directly provided in concentration units; all measured values are stored with date, time and sample O.D.; the same data are remotely available through a RS-232 serial communication port.

Reagents consumption is limited to about 50-60 µl of each reagent per single analysis (PO_4, NO_2) and 225 µl (NH_3, NO_3); the hand-held reagents container is designed to contain up to 500 ml of reagents and standard solution, to ensure about 500 complete field analysis. Auto-calibration is performed using concentrated standard solutions, contained in the same reagents container.

All these particularities make the WIZ as the most adapted probe for use on an autonomous buoy, where available energy and space are limited.

In the reported test the WIZ probe has been integrated in the MOLA buoy using its power supply (Figure 8). Data transmission was performed using a GSM programmable modem to manage the probe and sending Short messages Service (SMS) every four hours (the same frequency chosen for the tests) to a second phone, automatically forwarding the data to a MySql database, accessible by the Internet through a browser based application. In the near future the probe will be plugged directly to a RS-232 serial port to the buoy data logger, in order to recover data using WIFI connection like for the others sensors, automatically storing data in the same Laboratory database.

A first thing to be headlined was the simplicity of deployment of the probe in the buoy, which took only a half day, after a preliminary test in lab. To avoid creation of biofouling in the sampling line, an online filter was added (Figure 9) with a copper wire around it.

The WIZ has been fixed on the buoy at 2 meters depth on two points to avoid any rotation of the probe.

Fig. 8 WIZ deployment on MOLA buoy

Fig. 9 On line filter with copper wire

III. RESULTS AND DISCUSSION

Analytical procedure

The WIZ nutrients in-situ probe was configured to measure automatically and unattended the nutrient parameters (NH_3-N, PO_4-P, NO_3-N, NO_2-N) in sea water using the analytical sequence and the standard wet chemical methods reported in [13]. The sampling frequency chosen was adapted to the amount of reagents and standards that were filling the flexible bags. A two weeks field test was planned with a four hours sampling frequency to run cycles of four nutrients measurements. Online working standards were respectively of 100µg/L for ammonia, 100µg/L for phosphates, 20µg/L for nitrites and 150µg/L for nitrates.

Field tests

Figure 10 shows the concentration nutrient trend provided by the probe from August 26th to September 3rd, 2009. Very low values were obtained, typical from this period of the season and of the oligotrophic water on Côte Vermeille area. The good stability of the PO_4 (range 8–20µg/l, median 13.5µg/l) and NO_2 (range 0-3.8µg/l, median 2.32µg/l) concentrations in the harbour is highlighted. Due to a strong storm during the night on August 27th, an increase in NH_3 and NO_3 average concentration was observed (up to 22.5µg/l for ammonia, with a median of 6.4µg/l and up to 7µg/l for nitrate, with a median of 0.5µg/l).

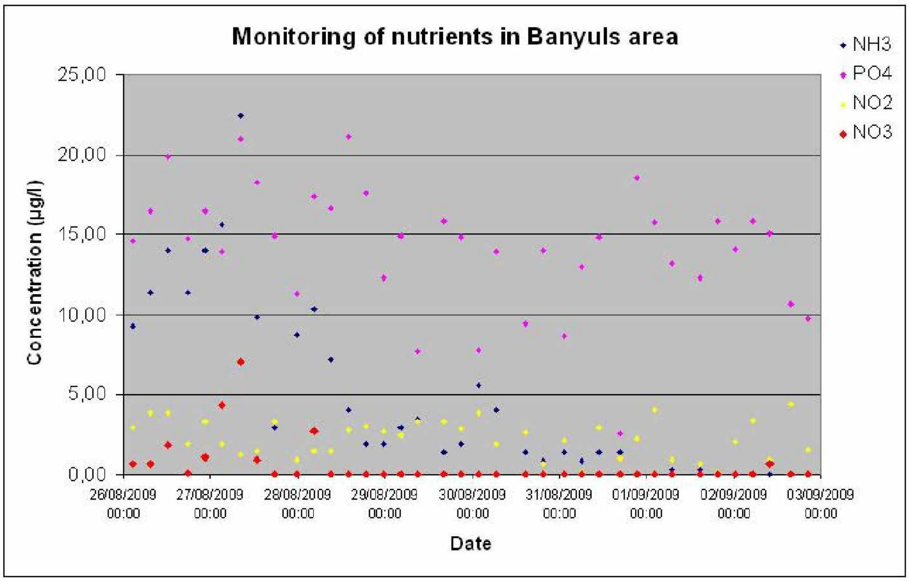

Fig. 10 Monitoring in the Banyuls harbour

The PO_4 and NO_2 data show the capability of WIZ probe to work on very low concentration, very closed to LOD with a good accuracy. The ammonia results were quite good during the first four days, after that a decrease down to zero values was noticed. Regular *in situ* calibration checks were made using on-board concentrated standard automatically diluted by the probe before the measurement; the results are shown in Figure 11. Even if no chloroform was placed in the internal standard to prevent calibrant degradation, a good stability for PO_4 and NO_3 was observed, with a small decrease of NO_2 during the first 3 days and than a substantial stability up to September 4th; NH_3 has been quite stable for the first 6 days and than it started to deteriorate.

After two weeks of deployment a visual check of the condition of the filter revealed that the protecting copper wire placed around the filter avoided the sample line to be clogged by biofouling.

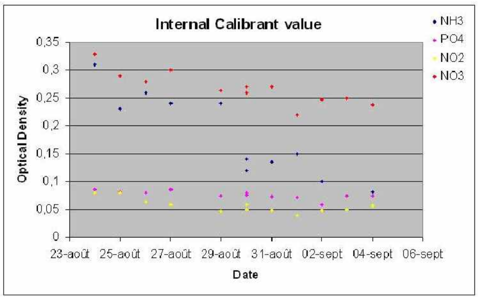

Fig. 11 Internal calibrant evolution

During the next deployments the internal concentrated standard solution will be prepared with higher ions concentration and Chloroform will be added to evaluate the stability and how long could be used the same standard *in situ* especially for ammonia.

IV. CONCLUSIONS

The deployment of the WIZ probe on *in situ* conditions in the Banyuls sur Mer harbour has been successfully performed with the demonstration of the capability of such system to work on an automated mode with a good accuracy. The probe deployed was loaded with reagents for 2 weeks autonomy and it carried sufficient quantity of standards and DI water to analyze four nutrient parameters continuously at 4h intervals during the reported period. The on-going experiments at the harbour show that the probe could be deployed for further weeks in a highly oligitrophic environment with minimal maintenance, as the copper wired filter avoided the creation of biofouling in the sample line.

The reduced power absorption requires less self generated power and permits a perfect adaptation to the MOLA buoy without perturbing the autonomy of the system. The compact design and the reagent container ensure a reduced weight and size and allow a quick, simple and easy reagents changeover, minimizing maintenance service on the buoy, performed by a single person.

Longer tests will be performed in order to have a better evaluation of the system especially on the accuracy and repeatability. A special effort has to be made on the standard solution for oligotrophic area; an intercomparison test with simultaneous sampling and analysis on lab is already planned.

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