UNIVERSITAT ROVIRA i VIRGILI

Facultat de Química Tarragona

# Evaluation of Nitrogen and Phosphorus removal in Tarragona Wastewater Treatment Plant: Impact of Reactor's Temperature and System Stability


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Primer de tot, vull agrair a la Carme Aguilar, la meva tutora acadèmica, qui ha dedicat hores i hores revisant totes les versions del treball, amb mil suggerències a comentar, sempre amb l'objectiu d'ajudar-me a buscar la millor versió de mi mateixa en la realització d'aquest treball. Potser al principi sí que és veritat que m'espantava cada cop que rebia les seves correccions, però ara, mirant enrere, només puc donar-li les gràcies per haver-me acompanyat durant tot aquest curs.

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# 1.ABSTRACT

This Final Degree Project aims to evaluate the nutrient removal efficiency of the Wastewater Treatment Plant (WWTP) of Tarragona, in order to assess whether it could comply with potential future regulations imposing stricter limits on phosphorus and nitrogen levels in the effluent. The study also investigates in detail how nutrient reduction occurs throughout different treatment stages, with a particular focus on the nitrification and denitrification processes taking place in the biological reactors.

The project begins with a contextualization of the importance of urban wastewater treatment, explaining its general process and the origin of these nutrients. It then describes how these nutrients —specifically, nitrogen in the forms of nitrite, nitrate, ammonium, and total nitrogen, and phosphorus as orthophosphate and total phosphorus— are removed during treatment. The experimental section outlines the sampling protocols, analytical methods used, and the instruments and equipment required to obtain empirical data. Furthermore, a possible correlation with MATLAB between the temperature of the biological reactor and nutrient removal efficiency in Tarragona's WWTP is explored, with the aim of determining whether nutrient reduction improves significantly during warmer months.

Additionally, the system's stability is evaluated through control charts for parameters such as suspended solids (SS) and biochemical oxygen demand (BOD5), illustrating performance trends from September 2024 to March 2025. These results are compared against the design removal efficiency of the plant established at a value of 90%. Chemical oxygen demand (COD) is also analyzed, and although no design removal rate is available for this parameter, the effluent values are plotted to confirm they are kept below the regulatory limit, indicating high process efficiency.

Finally, the conclusions highlight the system's consistency, demonstrated by its stable performance and effective nutrient removal in the evaluated period. Additionally, the assessment considers whether the Tarragona plant is adequately prepared to comply with potential future regulatory requirements.

*Aquest Treball de Fi de Grau té com a objectiu avaluar l'eficiència en l'eliminació de nutrients de l'Estació Depuradora d'Aigües Residuals (EDAR) de Tarragona, amb la finalitat de determinar si podria complir amb possibles futures normatives que imposin límits més estrictes en els nivells de fòsfor i nitrogen a l'efluent. L'estudi també investiga en detall com es produeix la reducció de nutrients al llarg de les diferents etapes del tractament, amb especial atenció als processos de nitrificació i desnitrificació que tenen lloc als reactors biològics.*

*El projecte comença amb una contextualització de la importància del tractament d'aigües residuals urbanes, explicant el procés general i l'origen d'aquests nutrients. A continuació, es descriu com s'elimina el nitrogen —en forma de nitrit, nitrat, amoni i nitrogen total— i el fòsfor —com a ortofosfat i fòsfor total— durant el tractament. La secció experimental detalla els protocols de mostreig, els mètodes analítics emprats i els instruments i equips necessaris per obtenir dades empíriques. A més, s'explora una possible correlació mitjançant MATLAB entre la temperatura del reactor biològic i l'eficiència en l'eliminació de nutrients a l'EDAR, amb l'objectiu de determinar si la reducció de nutrients millora significativament durant els mesos més càlids.*

*Addicionalment, s'avalua l'estabilitat del sistema mitjançant cartes de control per a paràmetres com els sòlids en suspensió (SS) i la demanda biològica d'oxigen a cinc dies ($DBO_5$), mostrant les tendències de rendiment des del setembre de 2024 fins al març de 2025. Aquests resultats es comparen amb l'eficiència de disseny, fixada en un 90%. També s'analitza la demanda química d'oxigen (DQO), i encara que no es disposa d'un valor de disseny per a la seva eliminació, els valors de l'efluent es representen per confirmar que es mantenen clarament per sota del límit normatiu, indicant una alta eficiència del procés.*

*Finalment, les conclusions posen de manifest la consistència del sistema, demostrada pel seu rendiment estable i la seva capacitat d'eliminació de nutrients en el període evaluat. A més, s'avalua si la planta de Tarragona està adequadament preparada per complir amb possibles futurs requisits normatius.*

# 2.INTRODUCTION

Ematsa (E*mpresa Mixta d'Aigües de Tarragona, S.A.*) is responsible for managing the integral water cycle in Tarragona and its surrounding municipalities. Its operations include water collection, distribution, as well as the maintenance of the sewage network and wastewater treatment.

In the field of sanitation, Ematsa operates several wastewater treatment plants (WWTPs), where a combination of physical, chemical, and biological processes is applied to remove pollutants. Once treated, the effluent is safely returned back to the natural environment under safe conditions. These treatment processes are essential to protect both the environment and public health, as well as to ensure the sustainable use of water resources.

## 2.1 Importance of Wastewater Treatment

Wastewater treatment is one of the most relevant activities of environmental management due to its direct impact on both human health and the stability of aquatic ecosystems. Municipal wastewater from households and urban areas contains a wide range of pollutants, with nitrogen and phosphorus being particularly significant due to their environmental impact[1]. If these nutrients are not properly removed, they can lead to eutrophication processes in aquatic environments, resulting in oxygen depletion and excessive algae growth, which in turn negatively affects water quality and aquatic biodiversity[2].

The presence of high concentrations of nitrogen compounds (such as ammonia, nitrites, and nitrates) can cause environmental risks, including toxicity to aquatic life, decreased oxygen levels, and potential risks to human health. Similarly, high phosphorus concentrations act as a limiting nutrient in freshwater ecosystems, triggering rapid algal blooms. These blooms not only reduce water transparency and aesthetic value but may also produce toxic substances harmful to both humans and animals.

To prevent such impacts and ensure environmental safety, it is essential to have a system which efficiently purifies the wastewater. The next section outlines the purification system of Tarragona's WWTP.

## 2.2 Purification System

The Tarragona sanitation system treats wastewater from the city of Tarragona, the Francolí industrial hub, and the Riu Clar industrial hub. The wastewater management system operates mainly through two pumping stations: *Francolí* and *Entrevies*, which transfer water directly to the WWTP. The *Francolí* station also receives water from several subsidiary stations, including *Serrallo*, *Bonavista*, *Miracle*, and *Barcelona*. Upon treatment, the effluent is either discharged into the sea through the *Riera de Riu Clar* or pumped to the Vilaseca-Salou WWTP for further tertiary treatment.

The Tarragona's WWTP operates at an average flowrate of 24000 $m^3$/day, even though its maximum design capacity is 35000 $m^3$/day. The treatment system consists of two main processing lines:

1. Water Line – consisting of pre-treatment, primary treatment, and secondary treatment stages.
2. Sludge Line – includes thickeners, anaerobic digestion, and sludge dewatering.

The figure below shows the schematic diagram of Tarragona's wastewater treatment plant, illustrating the complete process with all the stages involved for both treatment lines.

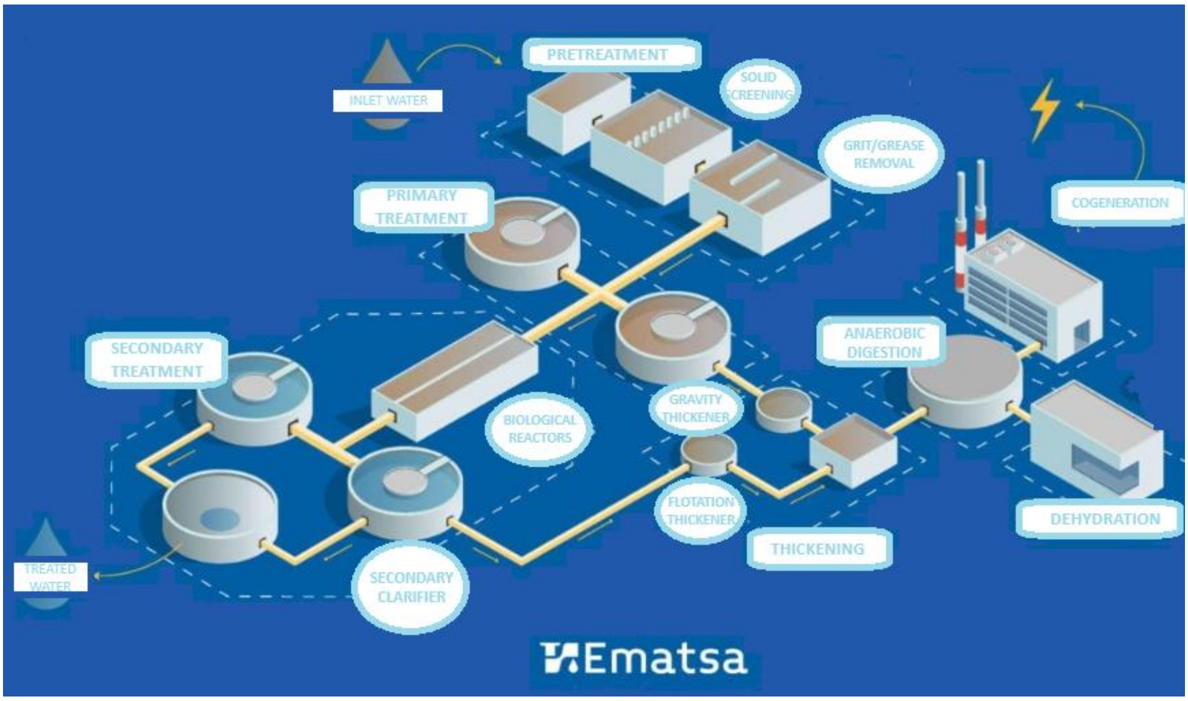


*Figure 1. Flow diagram of Tarragona's WWTP with all stages of the purification process for both the water and sludge lines.*

As illustrated in the figure, the process begins with raw wastewater pretreatment, followed by primary and secondary treatments to produce purified effluent. Simultaneously, the generated sludge undergoes a thickening process and anaerobic digestion and ends with dehydration for its potential subsequent agricultural use.

In the following sections, the two lines that constitute the overall purification process are explained in detail.

### 2.2.1 Water line

The water line consists of three main stages designed to progressively improve the water quality. In particular, these stages are:

- **Pre-treatment:** involves screening using 5mm and 3mm sieves for solid separation, followed by grit and grease removal using air injection and gravitational separation.
- **Primary treatment:** uses clarifiers equipped with rotating bridges to remove settled sludge and floating materials.
- **Secondary treatment:** occurs in biological reactors (with a total volume of 3,697 $m^3$) where microorganisms degrade organic matter over a hydraulic retention time of roughly 8 hours, followed by secondary clarification which allows biomass to settle. A portion of the activated sludge is recirculated to maintain microbial populations, while excess sludge from both clarification stages is directed to the sludge treatment line for further treatment.

### 2.2.2 Sludge line

The sludge treatment involves several steps:

- **Thickening**: primary sludge is thickened by gravity settling, while secondary sludge is thickened via flotation using pressurized water.
- **Anaerobic Digestion:** both thickened sludges are combined in a mixing chamber before entering the anaerobic digestor where organic matter undergoes four biological stages (hydrolysis, acidogenesis, acetogenesis, and methanogenesis) to produce biogas and inert sludge. The biogas is used on-site for heating and electricity generation for the plant, contributing to the plant's energy self-sufficiency.
- **Dewatering**: the digested sludge is dehydrated through centrifugation to achieve high dry matter content, making it suitable for agricultural application (e.g., as fertilizer). Throughout the process, separated water is recirculated back to the plant's influent for reprocessing.

## 2.3 Essential Nutrients and their Removal

To mitigate eutrophication risks, WWTPs must effectively reduce nutrient levels, particularly nitrogen and phosphorus, however, many conventional WWTPs are not specifically designed for the efficient removal of nutrients. Consequently, there is a growing demand for improved treatment strategies to enhance nutrient removal, particularly in anticipation of potential future regulations which may impose stricter limits on effluent water quality.

High concentrations of nitrogen (in the forms of nitrate, nitrite, or ammonia) and phosphorus in the effluent discharges can lead to significant environmental problems as it has been mentioned previously. Reducing or eliminating these nutrients during the treatment process is crucial to prevent these negative impacts, ensuring the protection of ecosystems, maintaining water quality, and safeguarding public health.

Tarragona WWTP, is an example of how a facility not being originally designed for the efficient removal of these nutrients, has successfully achieved a reduction in nitrogen and phosphorus levels in its effluent waters. This highlights the importance of understanding the specific behavior of nutrients within the treatment process, as they represent a growing challenge in wastewater treatment.

The following subsections describe the sources, chemical forms and the available mechanisms for the removal of these two key nutrients.

### 2.3.1 Nitrogen: sources and removal

Nitrogen in wastewater typically appears in four main forms: ammonium ($NH_4^+$), nitrate ($NO_3^-$), nitrite ($NO_2^-$) and organic nitrogen. These forms primarily originate from a variety of sources as[3]:

1. Industrial wastewater – Sectors such as agri-food, textile, and chemical industries often discharge significant amounts of organic nitrogen and ammonium into wastewater.
2. Domestic wastewater – Human urine contains high concentrations of urea ($CO(NH_2)_2$), which hydrolyzes into ammonium ions ($NH_4^+$). Additionally, feces also contribute organic nitrogen in the form of proteins and amino acids. Moreover, some household cleaning products and detergents can also contain nitrogen-based compounds.
3. Stormwater runoff – Urban runoff transports nitrogen-based compounds (nitrates, nitrites, ammonium) from fertilizers and atmospheric deposition.

At Tarragona's WWTP, the removal of nitrogen in biological reactors is based on the nitrification-denitrification (NDN) process[4.] This is a biological two-steps process, which involves:

1. **Nitrification**: this aerobic process is carried out through two steps by the *Nitrosomas* bacteria:

A) Step 1. Ammonium is oxidized to nitrite by ammonium-oxidizing bacteria (1).

$$(1)\ 2NH_4^+ + 3O_2 \rightarrow 2NO_2^- + 2H_2O + 4H^+ + energy$$

B) Step 2. Nitrite is further oxidized to nitrate (2) by nitrite-oxidizing bacteria.

$$(2)\quad 2NO_2^- + O_2 \rightarrow 2NO_3^- + energy$$

2. **Denitrification**: this anoxic process is performed mainly by *Pseudomonas* bacteria which are responsible for removing nitrates ($NO_3^-$) and converting to nitrogen gas ($N_2$). This step involves the reduction of nitrates to nitrites (3), then to nitric oxide, then to nitrous oxide, and finally to nitrogen gas.

$$(3)\ NO_3^- \rightarrow NO_2^- \rightarrow NO + N2O \rightarrow N_2$$

In the following figure, there is a simplified diagram illustrating these stages

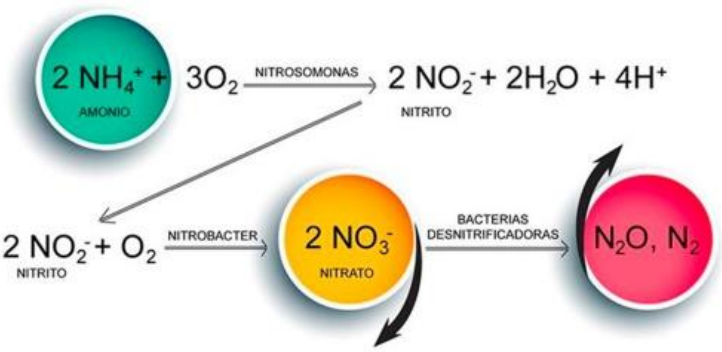


*Figure 2. Simplified representation of the nitrogen cycle, illustrating the main transformation stages and chemical forms involved in the nitrogen removal during wastewater treatment.*

The nitrogen cycle in wastewater treatment involves specific steps such as the aerobic oxidation of ammonium to nitrite (by ammonium-oxidizing bacteria), the subsequent aerobic oxidation of nitrite to nitrate (by nitrite-oxidizing bacteria) during nitrification, and the anoxic reduction of nitrate and nitrite to dinitrogen gas via denitrification, which enables partial or total nitrogen removal from the system.

### 2.3.2 Phosphorus: sources and removal

Phosphorus compounds in wastewater primarily come from fertilizers via runoff, human and animal excretions, detergents and cleaning products used in households and industry[5-7].

The total phosphate load (TP) in wastewater is composed of three main forms: orthophosphate (P-$PO_4$), polyphosphate (Poly-P) and organic phosphorus compounds ($P_{org}$).

Phosphorus removal can occur through two main processes:

1. **Biological removal process**. Involves phosphorus accumulating organisms (PAOs) in two stages:

   - Anaerobic stage: PAOs absorb volatile fatty acids (VFAs) and release phosphorus.

   - Aerobic stage: PAOs degrade the organic matter they previously absorbed and the consequent energy they generate for their growth is reused to capture the phosphorus. This process allows the elimination of phosphorus by extracting the sludge generated[6].

2. **Chemical removal process**. Involves adding coagulants as $FeCl_3$, which react with phosphate to form insoluble ferric phosphate, and then this can be removed from water[8].

$$(4)\ FeCl_3 + PO_4^{3-} \rightarrow FePO_4 \downarrow + 3Cl^-$$

While both methods are effective, Tarragona's WWTP primarily relies on biological removal. Although $FeCl_3$ is added in the mixing chamber, it is not specifically intended for phosphorus reduction, but rather for odor control and coagulation, which facilitates better sludge compaction during dewatering.

## 2.4 Effect of Operational Conditions on Biological Processes

The efficiency in nutrient removal in wastewater plants depends heavily on the activity and performance of microbial communities. These biological processes are highly sensitive to different operational parameters, among which the temperature, dissolved oxygen or hydraulic retention time are significantly important.

The following subsections present two key aspects that serve as the foundation and motivation for this in-depth study: preliminary observations from an internal investigation and the influence of biological reactor temperature on the treatment process.

### 2.4.1 Preliminary Observations from Microbial Analysis

A recent internal metagenomic study conducted on the biological reactors at the Tarragona WWTP provided valuable insights into the microbial composition of the system. This study, although unpublished and restricted exclusively for internal use, applied high-throughput genetic sequencing techniques to identify and quantify the microbial communities present in the treatment process.

The analysis identified over 2.700 microbial species, with a clear prevalence of phosphorus-accumulating organisms (PAOs), which play a key role in biological phosphorus removal[9]. That study was conducted by Dr. Andrés Zornoza Zornoza, a leading expert in microscopy and wastewater microbiology. As co-founder of WALEBUBLE, Dr. Zornoza has made significant contributions to the field, particularly in the identification and optimization of microbial communities to improve the performance of water treatment processes.

The findings of that study demonstrated that PAOs were the most abundant microbial group, suggesting their crucial role in the phosphorus reduction. The relevance of this study lies not only in confirming the dominance of PAOs, but also in highlighting its potential to inform operational strategies through the integration of molecular microbiology tools into routine monitoring, thereby ensuring the effectiveness of the purification process. A thorough understanding of the microbial community enables plant operators to anticipate performance fluctuations and implement proactive measures to optimize system stability and efficiency, especially in the face of variable influent characteristics or environmental conditions.

Moreover, this preliminary microbial analysis confirms the key role of PAOs in the current treatment process and highlights the importance of microbiological monitoring to prepare for possible future regulations.

### 2.4.2 Influence of Temperature on the Nutrient Removal Efficiency

Among the various operational parameters in wastewater treatment, the temperature of the biological reactor plays a key role in the reduction of nutrients such as nitrogen and phosphorus. Changes in temperature throughout the year can significantly influence the metabolism of bacteria present in the reactors, impacting the efficiency of nitrogen and phosphorus reduction[10].

Previous studies have demonstrated that at lower temperatures, microbial growth and metabolic activity tend to slow down dramatically, potentially leading to partial inhibition of biological treatment processes. Conversely, warmer conditions enhance microbial activity and may improve nutrient removal efficiency[11].

To investigate and better understand the influence of this parameter at Tarragona's WWTP, this bachelor thesis includes a statistical analysis of historical operational data related to nutrient removal using MATLAB. The main aim is to determine whether there is a noticeable improvement in nutrient reduction during periods of rising temperatures, when microbial activity is expected to be enhanced.

In addition to evaluating nutrient removal efficiency, it is essential to assess whether the treatment system is capable of maintaining consistent performance over time[11,12]. These articles highlight the importance of understanding the parameters evaluated and their removal efficiencies and mention how the system's stability is considered a key for reliability and efficient performance. Basically, the studies are based on the observation of the correlation between a good stability and a more efficient yield.

Stability in core parameters, such as $BOD_5$ and suspended solids, is widely considered as a fundamental prerequisite for ensuring reliable robustness and system efficiency, especially under varying operational conditions[13].

For this reason, a dedicated section of the results focuses on assessing system stability through the use of control charts and performance trend analysis. This assessment provides a necessary foundation for interpreting the nutrient removal data and for evaluating the plant's preparedness to meet increasingly stringent regulatory requirements. Moreover, understanding the system's ability to maintain stable operating conditions is crucial for the early detection of potential process disturbances and for the implementation of proactive corrective actions. By systematically analyzing historical data, it becomes possible to identify patterns, seasonal variations, and operational challenges that may affect long-term system performance.

Previous studies have underscored that system stability is not merely desirable but essential to ensure the robustness and resilience of wastewater treatment operations[13,14]. An adequate stability guarantees environmental security, minimizing noncompliance with current regulations as well as understanding the biological treatment process can optimize energy consumption.

## 2.5 Regulatory Considerations

At the Tarragona's WWTP, treated effluent is discharged into a public watercourse in compliance with the current wastewater regulations including Directive 91/271/EEC[15], Royal Decree 509/1996[16] (March 15), and Decree 130/2003[17] (May 13, D.O.G.C 3894), which establish wastewater treatment regulations at European, national and regional levels, respectively.

The table below summarizes the maximum allowed limits for discharge into public watercourses applicable to urban wastewater treatment plants. These limits are established by the Royal Decree 509/1996.

*Table 1. Current Maximum Allowed Limits for Discharge for urban WWTP effluent (RD 509/1996)*

| Parameter | Maximum Allowed Limits |
|---|---|
| **COD** (Chemical Oxygen Demand) | 125 mg/L |
| **$BOD_5$** (Biochemical Oxygen Demand) | 25 mg/L |
| **SS** (Suspended Solids) | 35 mg/L |

Although current regulations do not explicitly mandate specific nutrient removal in WWTPs, the potential implementation of such standards in the near future could pose significant challenges for WWTPs not originally designed for this purpose. Complying with stricter nutrient discharge limits may require technically complex and costly facility modifications, or in some cases, complete redesigns of the treatment infrastructure. Interestingly, preliminary microbial and operational data suggest that the Tarragona WWTP already achieves a certain degree of nutrient removal, despite not being specifically designed for this purpose. This unexpected capability presents an opportunity to evaluate the plant's potential to meet future regulatory requirements without the need of major structural modifications.

# 3.OBJECTIVE

The main objective of this Bachelor Thesis is to analyze the behavior of nitrogen and phosphorus reduction within the biological treatment processes of the Tarragona's WWTP. This analysis is conducted with a view to the potential implementation of future stricter regulations regarding these nutrient levels in effluents. Additionally, this study aims to investigate the possible correlation between the temperature of the biological reactor (BR) and the efficiency in the removal of these nutrients, particularly considering seasonal variations recording data from February-25 to April-25.

A further objective is to demonstrate the operational stability of the treatment system over time by analyzing key performance indicators such as $BOD_5$ and SS. Ensuring that the plant maintains consistent treatment efficiency over time is essential to validate conclusions related to nutrient removal performance. To this end, analytical data were collected and evaluated through control charts over a representative period from September 2024 to March 2025.

By understanding the relationships between operational parameters, seasonal influences and treatment outcomes, this study aims to contribute to the optimization of nutrient removal processes and support compliance with both current and potentially stricter future regulations.

# 4. EXPERIMENTAL PART

This section describes the experimental methodology used to evaluate the nutrient removal performance of Tarragona WWTP. It includes a description of the sampling locations and frequency, and the reagents and the analytical methods used. It also provides the equipment and procedures used for the assays required for assessing the system's stability.

## 4.1 Sampling Locations and Frequency

To comprehensively assess nutrient removal throughout the treatment process, samples were collected at five strategic points within Tarragona's WWTP. These locations were selected to provide a complete overview of the influent water quality, the intermediate treatment performance, and the final effluent water quality.

*Table 2* summarizes the sampling points including a brief description of each, their location and its role within the treatment process.

*Table 2. Sampling locations and descriptions*

| Name Code | Description | Sampling Spot |
|---|---|---|
| IAE | Internal Influent Water | Grit and grease removal bridge's exit |
| IAD | Internal Settled Water | Primary clarifier's exit |
| IAS | Internal Effluent Water | Secondary clarifier's exit |
| IRB A | Internal Biological Reactor A | Tail end of Aeration tank A |
| IRB B | Internal Biological Reactor B | Tail end of Aeration tank B |

The influent, settled, and effluent water samples were collected as composite samples, using an automated sampler programmed the day prior to collection. Composite sampling in WWTP provides a more accurate representation of wastewater quality over a specific period of time, as it integrates multiple subsamples taken at regular intervals, thereby minimizing the influence of short-term fluctuations.

Regarding the biological reactor's samples, spot sampling was used. Automatic samplers are not recommended in biological reactors since its content is in constant agitation, so a spot sample is representative of the whole, making continuous sampling unnecessary. Additionally, mixed liquor suspended solids can clog the tubes and pumps of the automatic sampler, causing operational failures and requiring frequent maintenance.

Key operational parameters within the biological reactor such as dissolved oxygen, pH, and temperature are continuously monitored through in-line sensors, which are more appropriate for this point of the process, as they provide real-time data.

Sampling was conducted with a minimum frequency of twice per week. Depending on the workload and the availability of analytical kits, additional sampling days were occasionally added to collect more data.

For the purpose of constructing control charts to assess the system's stability, data have been compiled from operational records collected twice per week.

## 4.2 Reagents Used

The concentrations for nitrite, nitrate, ammonium, total nitrogen, orthophosphate and total phosphorus were determined by using ready-to-use tube tests designed for photometric analysis manufactured from Macherey-Nagel (Düren, Germany). These test kits are specifically designed for their use with spectrophotometers and allow for rapid, accurate quantification of nutrient concentrations in water samples. Each kit contains pre-dosed reagents for the specific nutrient to be determined.

*Table 3* summarizes the Macherey-Nagel kits used in this study, detailing their chemical content, as well as the associated hazards, and the recommended protective measures.

*Table 3. Kits for nutrient analysis and their safety considerations.*

| **Kit** | **Test Tube's content** | **Hazards** | **Protective Measures** |
|---|---|---|---|
| Nitrite (0.1–4.0 mg/L $NO_2$-N) | sulfanilamide and N-(1-naphthyl)-ethylenediamine | None | Lab coat and suitable gloves |
| Nitrate (0.3–22.0 mg/L $NO_3$-N) | 2,6 - Dimethylphenol |  | Lab coat, suitable gloves and safety goggles |
| Ammonium (1–40 mg/L $NH_4$-N) | Indophenol |  | Lab coat, suitable gloves and safety goggles |
| Total Nitrogen (0.5–22.0 mg/L N) | 2,6 - Dimethylphenol |  | Lab coat, suitable gloves and safety goggles |
| Total Phosphorus (0.30–15.00 mg/L P) | Molybdenum blue |  | Lab coat, suitable gloves and safety goggles |

*Table 4* lists all the reagents used in the analytical procedures related to the system stability assays. Each entry includes the assay type, the commercial brand, hazard classification, and the recommended protective measures.

*Table 4. Reagents and safety considerations for the evaluation of the system stability.*

| Reagents | Assay in which is used | Brand | Hazards | Protective Measures |
|---|---|---|---|---|
| Nitrification inhibitor (N-allylthiourea) | $BOD_5$ assay | EMATSA (prepared solution) | Non-Hazardous | Lab coat, safety goggles, suitable gloves |
| Sodium Hydroxide pellets (>98% purity) | $BOD_5$ assay | Merck (Darmstadt, Germany) |  | Lab coat, safety goggles, suitable gloves |
| COD Cell 100-1500 mg/L $O_2$ | COD influent water assay | Macherey Nagel |  | Lab coat, safety goggles, suitable gloves |
| COD Cell 15-160 mg/L $O_2$ | COD effluent water assay | Macherey Nagel |  | Lab coat, safety goggles, suitable gloves |

## 4.3 Materials and Equipment

The analytical kits used were provided with all reagents pre-loaded in the test-tubes, minimizing the need for additional materials. However, for samples where the nutrient concentrations exceeded the measurement range of the kits, they were diluted using volumetric flasks. Furthermore, micropipettes were used to accurately measure the required volumes for dilution.

The main equipment used throughout this study was the spectrophotometer Nanocolor 550D (Macherey Nagel, Düren, Germany). Additonal laboratory equipment used for supplementary analyses related to system stability is listed in *Table 5*.

*Table 5. Equipment used for Tarragona's WWTP system stability performance*

| Equipment | Brand |
|---|---|
| Analytical Balance PIONEER | OHaus (Parsippany, New Jersey, USA) |
| Stove CONTERM | JP Selecta (Barcelona, Catalonia, Spain) |
| Thermoreactor | Macherey-Nagel (Düren, Germany) |
| Dessicator | OHaus (Parsippany, New Jersey, USA) |
| OxiTop $BOD_5$ measuring heads | WTW (Weilheim, Germany) |
| Vacuum filtration equipment | - |

## 4.4 Analytical Procedures

This section provides a detailed explanation of the followed procedures for each assay performed to evaluate both nutrient removal and system stability at the Tarragona's WWTP.

### 4.4.1 Nitrite

Nitrite analysis is based on a colorimetric method in which sulfanilic acid reacts with the nitrite ions present in the sample to form a diazonium cation. In the presence of N-(1-naphthyl)-ethylenediamine, the diazonium compound produces an azo-dye with a characteristic pink coloration. The intensity of the coloration, photometrically measured at 540 nm, is directly proportional to the nitrite concentration.

This method follows the analytical principles outlined in ISO 6777[18] by using Macherey-Nagel test kits and involves the following steps:

1- Pipette 200 µL of the sample into the test tube.
2- Add one NanoFix R2 pill (1-naphtyl amine).
3- Close the test tube and shake vigorously.
4- After 15 minutes, measure the absorbance at 540 nm.

The following figure shows the scheme of the reaction involved in the quantification of nitrite:

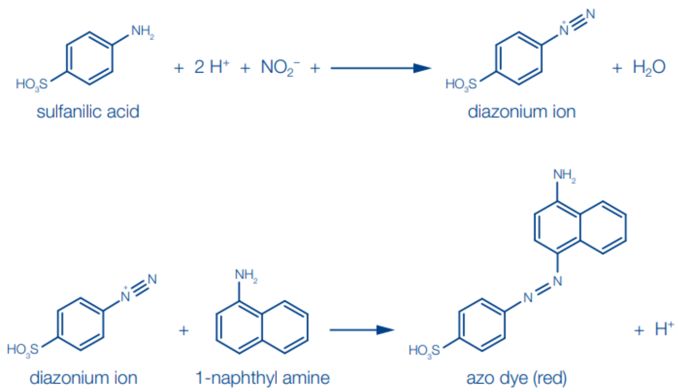


*Figure 3. Reaction scheme for nitrite detection using Macherey – Nagel Test Kits.*

### 4.4.2 Nitrate

The nitrate test relies on its reaction with 2,6-dimethylphenol in a mixture of sulfuric acid and phosphoric acid to produce 4-nitro-2,6-dimethylphenol, which is measured photometrically. The final color of the solution is pinkish (as in the nitrite test) and its intensity is directly proportional to the nitrate concentration. *Figure 4* represents the reaction involved in the quantification for nitrate.

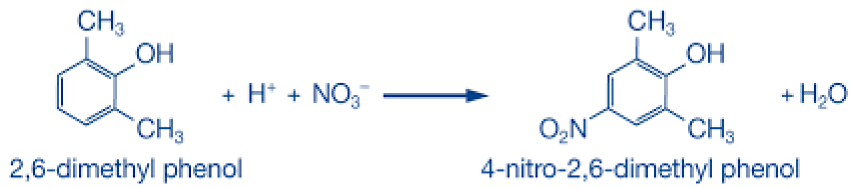


Figure 4. *Reaction scheme for nitrite detection using Macherey – Nagel Test Kits.*

The procedure is based on ISO 23696-1 2023-02[19] and briefly consists in:

1- Pipette 0.5 mL of the sample into the test tube.
2- Add 0.5 mL of R2 (2,6-dimethylphenol).
3- Seal tightly and shake vigorously.
4- After 10 minutes, measure the absorbance at 345/350/365 nm.

### 4.4.3 Ammonium

Ammonium ions ($NH_4^+$) or ammonia react with hypochlorite ($ClO^-$), which oxidizes the ammonium partially to monochloramine ($NH_2Cl$) according to the following reaction (5):

$$(5)\ NH_3 + NaOCl \rightarrow NH_2Cl + NaOH$$

This intermediate then reacts with salicylate to produce a blue-green indophenol compound, whose intensity is proportional to the concentration of ammonium in the sample. This compound is measured photometrically at 660/690 nm. The followed method is based on ISO 23695 2023-02[20] and briefly consists in:

1- Add 0.2 mL of the sample into the test tube measured with a micropipette.
2- Add NanoFix R2 pill (sodium salycilate).
3- Close and mix vigorously.
4- After 15 minutes, measure the absorbance at 660/690 nm.

*Figure 5* illustrates the reaction scheme involved in the quantification of ammonium.

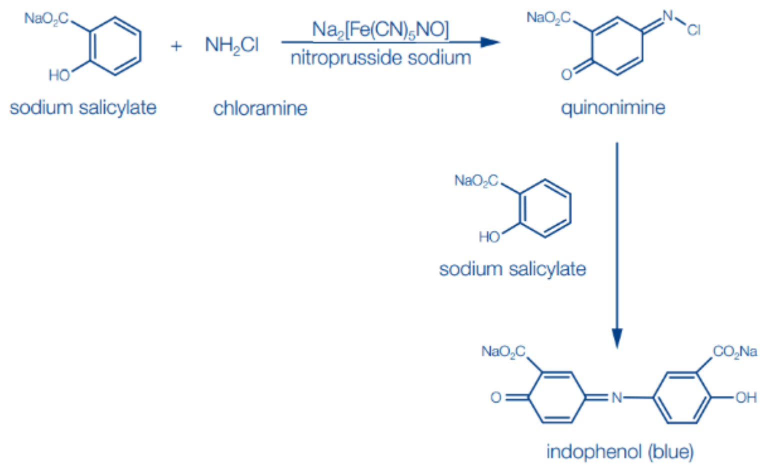


Figure 5. *Reaction scheme for ammonium detection using Macherey – Nagel Test Kits.*

The indophenol complex is formed through a two-step mechanism involving a quinonimine intermediate which subsequently reacts with sodium salcylate.

### 4.4.4 Total Nitrogen

According to ISO 23697-1 2023-02 total nitrogen determination involves the oxidation of all nitrogen-containing compounds (organic and inorganic) into nitrate using potassium persulfate ($K_2S_2O_8$), under heat[21]. The resulting nitrate ions then react with 2,6-dimethylphenol to form 4-nitro-2,6-dimethylphenol, a compound that can be quantified photometrically, this being last part of the previously described for nitrate determination in section 4.4.2.

The steps to follow are:

1- Add 5 mL of the sample to the digestion tube measured with a micropipette.
2- Seal and mix vigorously before placing it in the digestion block at 120ºC for 30 minutes.
3- Once cooled down, pipette 0.5 mL of this solution into a new test tube.
4- Add 0.5mL of R2 (2,6-dimethylphenol).
5- Shake and wait for 10 minutes.
6- Measure the absorbance at 345/350/365 nm.

The main oxidation reactions involved in the total nitrogen procedure are as follows:

$$(6)\ NH_4^+ + 2\ O_2 \rightarrow NO_3^- + H_2O + 2\ H^+$$

$$(7)\ NO_2^- + ½\ O_2 \rightarrow NO_3^-$$

### 4.4.5 Total Phosphorus and Orthophosphate

Polyphosphates—compounds in which phosphate units are linked by phosphorus–phosphorus bonds—and organic phosphates, which are bound to organic molecules, cannot react effectively with colorimetric reagents in their native forms. Therefore, an acidic digestion process is required to oxidize and hydrolyze these compounds into inorganic orthophosphate. This step ensures that all forms of phosphorus are converted into a measurable form, allowing for accurate and consistent quantification of total phosphorus.

After digestion, the orthophosphate reacts with ammonium molybdate under acidic conditions to form phosphomolybdic acid, which is subsequently reduced to a blue-colored molybdenum complex. The intensity of the resulting color is proportional to the phosphorus concentration and can be measured photometrically at 690 nm.

The main reactions taking place in the orthophosphate and total phosphate are as follows:

$$(8)\ PO_4^{3-} + 12\ MoO_4^{2-} + 24\ H^+ + 3\ NH_4^+ \rightarrow (NH_4)_3[P(Mo_3O_{10})_4] + 12\ H_2O$$

$$(9)\ (NH_4)_3[P(Mo_3O_{10})_4] + Na_2S_2O_5 \rightarrow \text{phosphorus molybdenum blue}$$

The steps to follow for the total phosphate are:

1. Pipette 500 µL of the sample into the test tube.
2. Add R2 (oxidant), close the test tube and mix.
3. Digest at 120ºC for 30 minutes.
4. After cooling down, add R3 (ammonium molybdate in acidic medium) to form phosphomolybdic acid.
5. Add 200 µL of R4 (sodium disulfite) to obtain the molydebnum blue complex.
6. Close the test tube, mix it vigorously and let it react for 10 minutes.
7. Measure the absorbance at 690 nm.

To determine orthophosphate only (without digestion), the procedure to follow is the same but omitting digestion (steps 2 and 3). The method follows the principles established in DIN EN ISO 6878-D11[22].

### 4.4.6 Suspended Solids (SS)

This assay quantifies the total suspended matter present in aqueous samples from influent water and effluent water samples.

In brief, SS are determined gravimetrically by the following procedure:

1. Place a pre-dried and weighted filter in a clean Petri dish.
2. Filter a known volume of the water sample (roughly 15 mL) using vacuum filtration.
3. Dry the filter in the oven at 105ºC for 24 hours.
4. Cool in a desiccator and weight.
5. Calculate the mass of retained solids and express as mg/L using the filtered sample volume.

### 4.4.7 Biochemical Oxygen Demand ($BOD_5$)

$BDO_5$ measures the amount of dissolved oxygen consumed by aerobic bacteria while breaking down organic matter over five days. The procedure consists in the following steps:

1. Measure specified volumes (97 mL for influent and 432 mL for effluent water samples) using volumetric flasks.
2. Add the sample to a BOD bottle with a magnetic stir bar.
3. Add the appropriate number of nitrification inhibitor drops as defined in protocol (N-allylthiourea solution) to the BOD bottle (2 and 9 drops for influent and effluent water samples, respectively).
4. Place a cap in the bottle and add one or two NaOH pellets.
5. Seal the bottle with an OxiTop head and incubate for five days at 20ºC.
6. Record the five OxiTop measurements (one for each day of incubation). To calculate the final $BOD_5$ value, multiply the last measurement (day 5) by the corresponding dilution factor: 20 for influent samples and 1 for effluent samples.

### 4.4.8 Chemical Oxygen Demand (COD)

COD assay measures the amount of oxygen required to oxidize both organic and inorganic matter in water by microorganisms. This analysis is performed on influent and effluent water samples and involves the presence of a strong oxidant which is potassium dichromate ($K_2Cr_2O_7$) in the case of Ematsa.

The corresponding procedure is as follows:

1. Pipette 2 mL of the homogenized sample into the COD test tube.
2. Seal the tube test and vigorously mix the content.
3. Digest the sample at 148ºC for 2 hours.
4. Allow the tube to cool at room temperature.

5. Measure the absorbance at 590 / 595 / 605 / 620 nm depending on the sample range and kit specifications in the spectrophotometer.

# 5.RESULTS AND DISCUSSION

This section presents and discusses the main results obtained from the analyses conducted during my dual mention internship at Ematsa. The primary focus is on studying nutrient removal efficiency in Tarragona's WWTP, with particular attention to the possible correlation between the efficiency of the nutrient reduction and the biological reactor's temperature. Additionally, system performance is evaluated through different operational parameters to ensure the stability of the system and to verify if the plant consistently meets its design requirements over a period of several months.

## 5.1 System Stability

The stability of a WWTP refers to the system's ability to consistently maintain effluent quality within the permitted or desired limits, despite variations in influent conditions (such as variations in flow rate, pollutant load, or temperature) or internal operational changes or adjustments.

To evaluate the stability of Tarragona's WWTP system, two key parameters were selected: $BOD_5$ and SS, as both directly relate to pollutant load and are essential for evaluating the plant's design performance. According to the facility's design documentation, the target removal efficiency for both parameters is 90%. Additionally, COD was included as a complementary parameter, although no specific target removals are established for this parameter because it offers a broader measure of total organic matter load while $BOD_5$ only represents the biodegradable fraction. Hence, the contaminant load can be evaluated in depth using the COD and $BDO_5$ results.

The data analyzed in the present study covers the period from September 2024 to March 2025. The typical concentration ranges obtained for these parameters in influent water in Tarragona's WWTP during the evaluated period are summarized in *Table 6*.

*Table 6. Expected concentration intervals of COD, SS and $BOD_5$ in influent water samples*

| Parameter | Concentration intervals |
|---|---|
| COD | 500-700 mg $O_2$/L |
| $DBO_5$ | 300-450 mg $O_2$/L |
| SS | 250-400 ppm |

The following subsections present and discuss the results obtained for each of these key parameters in more detail.

### 5.1.1 Biochemical Oxygen Demand

To evaluate $BOD_5$, monthly monitoring was conducted, and removal efficiencies were calculated to determine both the stability and the performance of the treatment process. The results are presented in *Table 7*, in which the average monthly values for influent and effluent $BOD_5$ concentrations, along with treated water volumes and the corresponding calculated removal efficiencies from September 2024 to March 2025, are included.

*Table 7. Monthly average $BOD_5$ levels for influent and effluent water samples from September 2024 to March 2025 and corresponding removal efficiencies*

| Month | Treated water ($m^3$/day) | Influent $BOD_5$ (mg $O_2$/L) | Effluent $BOD_5$(mg $O_2$/L) | Removal efficiency (%) |
|---|---|---|---|---|
| September-24 | 25226.5 | 335.0 | 12.9 | 95.7 |
| October-24 | 28808.5 | 327.5 | 9.9 | 96.6 |
| November-24 | 33996.8 | 227.5 | 13.5 | 93.8 |
| December-24 | 30241.8 | 342.2 | 10.4 | 96.7 |
| January-25 | 26054.6 | 455.0 | 18.6 | 95.5 |
| February-25 | 25441.9 | 460.0 | 16.9 | 96.1 |
| March-25 | 31205.2 | 380.0 | 13.6 | 96.1 |

As shown in the table, the results confirm the high and consistent $BOD_5$ removal efficiency performance of the WWTP throughout the entire monitoring period. Removal efficiencies remained all above 90% which is the design target value of the plant, highlighting its good performance. Only minor variations occurred, as the slightly lower efficiency in November (93.8%), likely due to Storm Dana, which increased the incoming water volumes, and consequently, diluted influent $BOD_5$ concentrations (227.5 ppm) compared to the usual values (300-450 mg $O_2$/L).

In contrast, during January and February, months with relatively dry weather, the treated water volumes returned closer to the usual range. As a result, the influent $BOD_5$ values also approached to the more typical levels, reflecting the standard performance of the plant.

To complement this initial evaluation over time, a control chart was constructed to assess day-to-day both the process stability and the variability in $BOD_5$ removal efficiency. *Figure 6* shows this control chart, with the 90% design target indicated for reference (in a blue line).

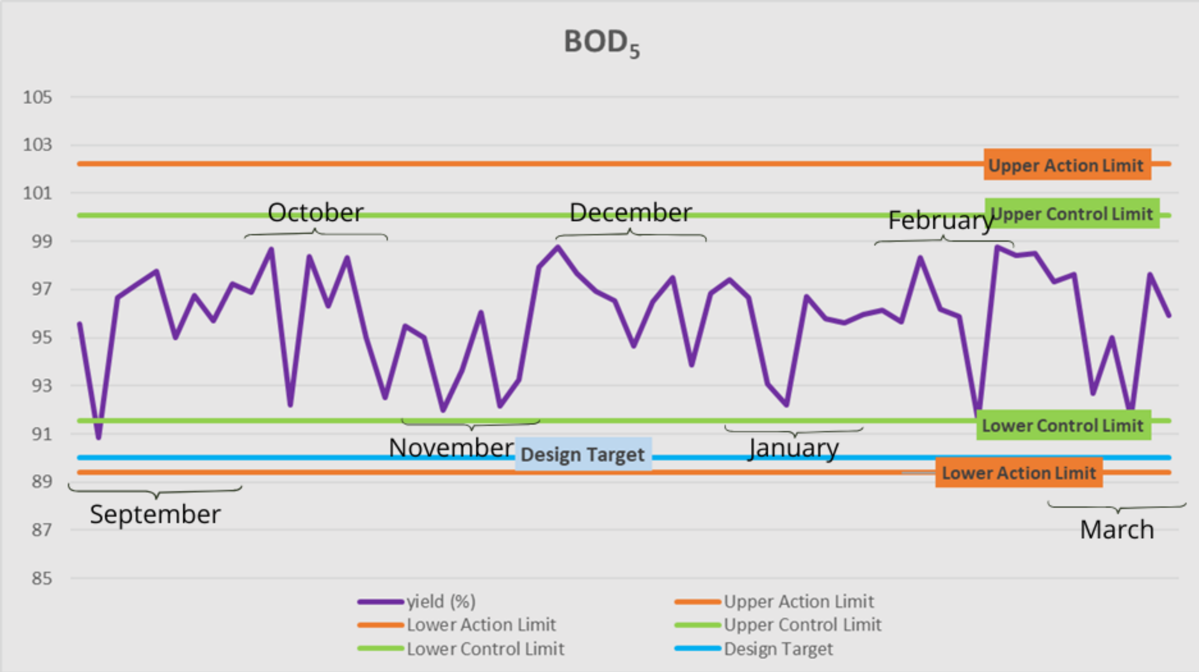


*Figure 6. Daily $BOD_5$ removal efficiency from September-24 to March-25 at Tarragona's WWTP*

As shown in the chart, $BOD_5$ removal efficiencies are significantly above the 90% design target, indicating that the WWTP system is very efficient in removing biodegradable organic load. These high removal percentages are indicative of strong biological activity in the system, as $BOD_5$ is highly sensitive to microbial activity in biological treatment processes. An efficiency above the design target efficiency may also suggest that the system has surplus treatment capacity, which could be due to favorable operating conditions or efficient biological treatment.

Exceptionally, only one point is slightly below the lower control limit (early September) and this was likely due to a particularly low influent $BOD_5$ (120 ppm on $5^{th}$ September) or a possible measurement error during my first week of the internship period. Nevertheless, system performance remained robust and within acceptable values. No trends indicating process instability were observed during the monitoring period, confirming that the WWTP operates reliably and effectively over that period, with removal efficiencies above 93% throughout the entire period under study. So, these results confirm that the Tarragona WWTP met and even exceeded its design target value in terms of $BOD_5$ reduction.

### 5.1.2 Suspended Solids

SS are a crucial indicator in wastewater treatment, as their poor removal can directly affect the quality of the treated water. Their effective removal ensures compliance with regulatory limits, and it is essential in assuring that the final effluent meets environmental and public health standards.

The optimal operation of the plant, based on its design performance, aims for a 90% reduction in SS. The following table provides monthly average SS concentrations from September 2024 to March 2025 for both influent and effluent water samples along with the calculated removal efficiencies.

*Table 8. Monthly SS concentrations for influent and effluent water samples and the corresponding removal efficiencies from September-2024 to March-2025*

| Month | Influent SS (ppm) | Effluent SS (ppm) | Removal efficiency (%) |
|---|---|---|---|
| September-24 | 318.0 | 20.3 | 85.5 |
| October-24 | 220.8 | 20.5 | 87.8 |
| November-24 | 172.4 | 18.1 | 88.0 |
| December-24 | 258.6 | 21.4 | 90.8 |
| January-25 | 398.3 | 20.2 | 93.7 |
| February-25 | 475.8 | 20.8 | 94.8 |
| March-25 | 387.0 | 21.6 | 93.0 |

As shown in the table the data collected show that the target value was not consistently reached every month, although the removal efficiency remains very close to the target. Not achieving the 90% does not mean that the plant is operating poorly, as the effluent values confirm that the facility consistently complies with current regulatory standards, ensuring the treated water is suitable for potential reuse or safe discharge into the environment.

These results can be linked to periods of increased flow and heavy rainfall, particularly in November due to the DANA storm, which caused significant dilution of the influent SS levels. For instance, influent SS concentration dropped to 172.4 ppm in November, and 220.8 ppm in October, both below the typical influent range of 250-400 ppm. Conversely, in months where no heavy rainfall occurred like January or February, the values for influent SS are within the expected range.

Unlike $BOD_5$, which dropped more markedly in the effluent because this parameter of the dilution effect caused by rainfall, SS levels at the effluent remained stable, consistently ranging between 18 and 20 ppm[23]. This explains why performance appeared slightly below design expectations: not because of insufficient treatment, but rather to lower than expected influent concentrations combined with consistently low effluent values. Overall, SS performance is more influenced by influent concentration, while $BOD_5$ responds more sharply to rainfall-driven changes.

To visualize the system stability regarding SS, a control chart in *Figure 7* was constructed, plotting the SS's removal efficiency across the monitoring period and comparing it to the 90% design target.

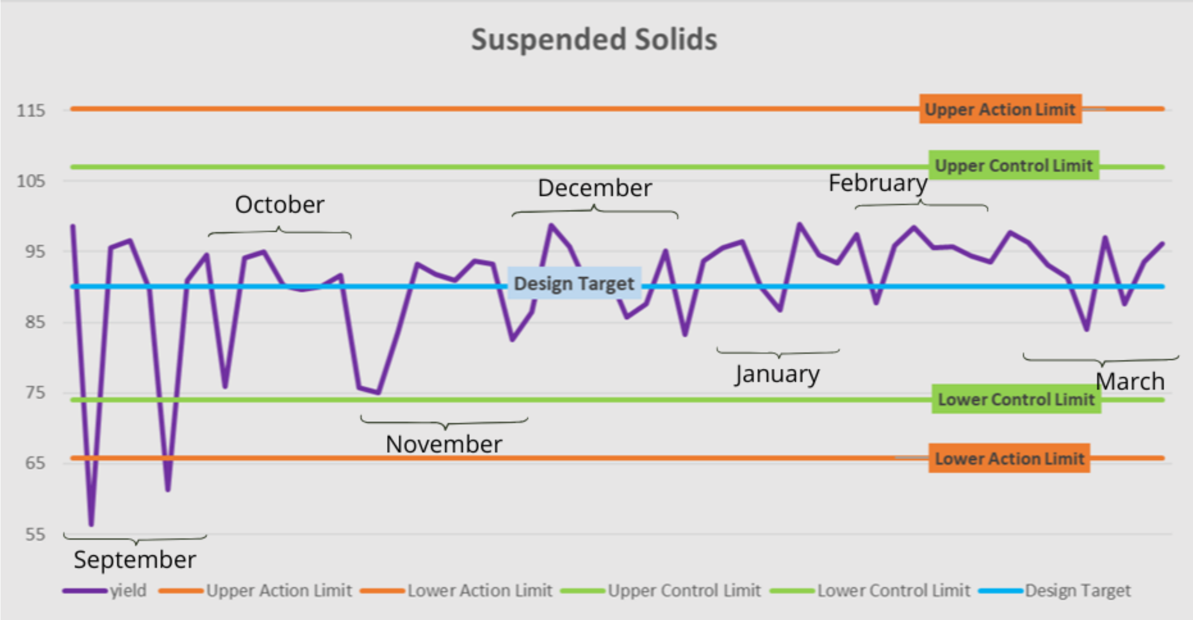


*Figure 7. SS removal efficiency from September-24 to March-25 in Tarragona's WWTP*

As shown in the chart, the majority of the data points are distributed close to or above the 90% target value and fall within the control and action limits, confirming that the system is working within stable conditions. Two isolated data points—September 5th and 19th—exceeded the lower action limit. However, these deviations do not indicate actual process instability, as on both days the influent SS concentration was unexpectedly low—around 80 ppm, compared to the usual 250–400 ppm range. There were no recorded changes in flow rate or operational parameters that could explain this drop. It is therefore that, exceptionally, the influent entered the plant with an unusually low influent suspended solids load. If the process had been genuinely unstable, other indicators such as effluent $BOD_5$ would also have shown similar anomalies. However, $BOD_5$ remained steady, confirming that overall performance was unaffected.

In summary, SS removal in Tarragona WWTP remained close or slightly above the design target throughout all the monitored period. The few deviations observed were due to unexpected and exceptionality of low SS contaminant load

### 5.1.3 Chemical Oxygen Demand

COD is an important parameter in wastewater treatment, representing the total amount of organic matter (both biodegradable and non-biodegradable) present in the water sample. It provides an indication related to the pollution load, and it is well used to evaluate the overall efficiency of the treatment process in reducing organic contaminants.

As for the previous parameters, to evaluate COD, monitoring was performed from September-24 to March-25 and the corresponding removal efficiencies were calculated.

*Table 9* summarizes the monthly averages of COD concentration levels in influent and effluent over the monitored period, along with calculated COD removal efficiencies.

*Table 9. Monthly COD levels for influent and effluent water samples and the corresponding removal efficiencies from September-24 to March-25*

| Month | Influent COD (mg $O_2$/L) | Effluent COD (mg $O_2$/L) | Removal Efficiency (%) |
|---|---|---|---|
| September-24 | 530 | 43 | 89.3 |
| October-24 | 568 | 61 | 88.6 |
| November-24 | 338 | 34 | 90.3 |
| December-24 | 589 | 36 | 93.5 |
| January-25 | 769 | 44 | 94.1 |
| February-25 | 771 | 63 | 91.5 |
| March-25 | 622 | 49 | 91.5 |

As shown in the table, the results demonstrate a high and consistent COD removal efficiency, between 88,6% and 94,1%. Despite the absence of a specific target value for COD removal efficiency, these values suggest effective organic load reduction in the evaluated period. Lower values in September (89.3%) and October (88.6%), which may be linked to the lowest influent COD levels. For these two months, the average values for COD in influent water was 530 mg $O_2$/L and 568 mg $O_2$/L, which were the lowest values reported in all the evaluated period, except for November (338 mg $O_2$/L) caused by the DANA storm, but despite this, removal efficiency reached 90.3% and effluent levels remained low (34 mg $O_2$/L) confirming the system adaptability to dilution events.

Higher removal efficiencies in January and February coincide with higher and more typical COD influent levels. Although rainfall continued into March, COD levels in both influent and effluent were more elevated than in November, likely due to occasional internal returns — recirculated flows from sludge treatment with high organic load. When such returns are detected, corrective actions are promptly implemented. Nevertheless, effluent values have consistently remained below the maximum permitted limits.

To assess how effluent values perform in regulatory terms, all COD measurements (from September-2024 to March-2025) were plotted in *Figure 8* together with the maximum discharge limit. Additionally, the graph displays the monthly average COD effluent value, further emphasizing the consistent performance of the treatment process. Note that removal efficiencies are not shown here, since there are no established design yields for COD in this treatment line and thus no baseline for comparison.

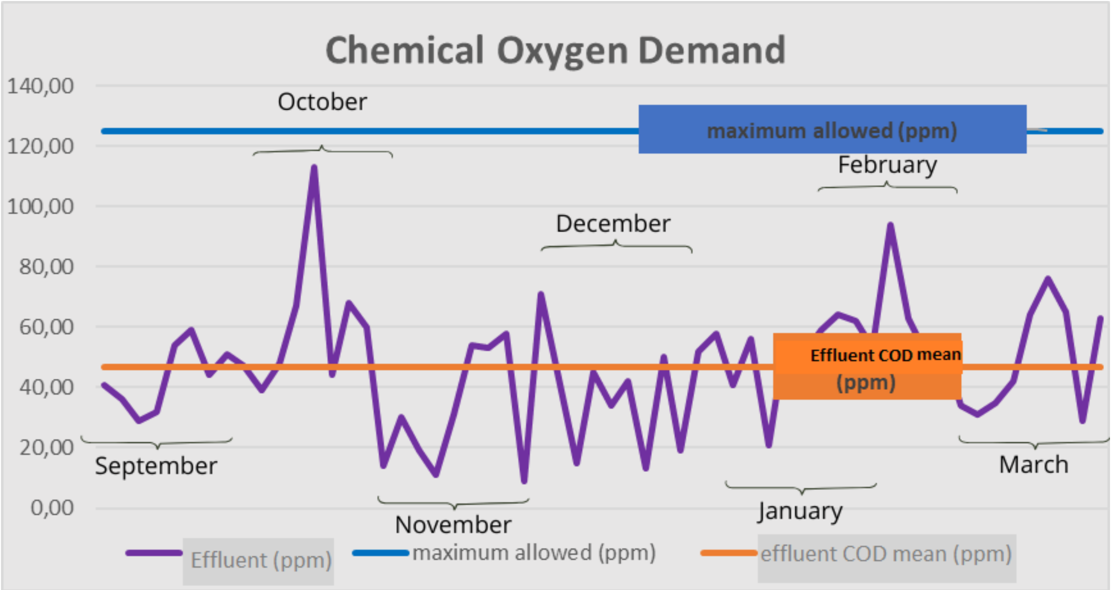


*Figure 8. COD effluent values from September-24 to March-25 in Tarragona's WWTP*

As shown in the figure, all measured effluent COD values remained below the maximum legal discharge limit (125 ppm), confirming that the treatment followed in the plant maintained full compliance during the entire period. Even under challenging conditions, such as abrupt change in influent flowrate or unexpected internal recirculation, the system worked under acceptable limits.

During months of heavy rainfall, such as November (monthly average of 33997 $m^3$/day) and December (monthly average of 30242 $m^3$/day), the plant experienced a decrease in the effluent COD levels. This is reflected in the graph, where a noticeable cluster of data points falls well below the average effluent COD. These fluctuations in water volume temporarily affect treatment efficiency, causing variability in the effluent results.

In contrast, the highest COD effluent concentration—113 ppm—was observed on October 17$^{th}$. Although this value edged closer to the regulatory threshold, it remained below. The spike may reflect either enhanced internal recirculation within the treatment process or an episodic industrial discharge (for example, from an industrial discharge or an unauthorized connection). Importantly, this anomaly was quickly addressed as the subsequent results returned to expected values.

Despite some variability and the occasional conditions caused by the rain, it's important to highlight that the effluent COD never exceeded the legal discharge limits in the studied period. This demonstrates the plant's ability to maintain low effluent COD concentrations despite seasonal variations and seasonal anomalies. So, the plant's ability to treat organic loads efficiently highlights the robustness of its design and of the treatment.

## 5.2 Nutrient Removal

This section presents and discusses the main results related to the removal of nitrogen and phosphorus conducted from February-25 to April-25. Both nutrients are key contributors to eutrophication and are expected to be subject to more stricter discharge regulations in the near future, established by European Directive 91/271/EEC[15]. Stricter levels on nitrogen and phosphorus are applied when zone is designated as sensitive due to eutrophication (<2 ppm TP and <15 ppm TN) Therefore, understanding the system's nutrient removal performance and its relationship with operational and seasonal factors is a key aim of the present study.

To evaluate how variables such as temperature and influent flowrates may influence in the concentration determinations, these parameters are summarized in the table below.

*Table 10. Monthly average temperatures and influent flowrate through the monitoring period*

| Month | Temperature (ºC) | Influent Flowrate ($m^3$/day) |
|---|---|---|
| **February-25** | 17.4 | 25749 |
| **March-25** | 17.8 | 29192 |
| **April-25** | 20.1 | 29644 |

As shown in the table, there is a marked increase in influent flowrate between February and March, likely reflecting seasonal rainfall during that period. Regarding temperature, the values span a relatively narrow range, indicating that seasonal temperature variations over this study period were moderate (from 17.4ºC and 20.1ºC).

### 5.2.1 Nitrogen

Nitrogen was monitored in the forms of nitrite, nitrate, ammonium, and total nitrogen. Concentrations were measured at five sampling points along the treatment process: influent water, settled water (after primary clarification), final effluent water and both biological reactors (A and B).

*Table 11* summarizes the monthly average concentrations of $NO_2$-N, $NO_3$-N and $NH_4$-N from February to April for influent, settled and effluent sampling points.

*Table 11. Monthly average concentrations of nitrogen species in influent, settled and effluent water samples over the monitored period.*

| | **Influent (ppm)** | | | **Settled (ppm)** | | | **Effluent (ppm)** | | |
|---|---|---|---|---|---|---|---|---|---|
| | $NO_2$-N | $NO_3$-N | $NH_4$-N | $NO_2$-N | $NO_3$-N | $NH_4$-N | $NO_2$-N | $NO_3$-N | $NH_4$-N |
| **February-25** | 0.2 | 0.6 | 48.8 | 0.1 | 0.6 | 46.2 | 0.6 | 1.3 | 39.1 |
| **March-25** | 0.2 | 0.7 | 45.1 | 0.1 | 0.5 | 41.2 | 1.0 | 2.8 | 18.3 |
| **April-25** | 0.1 | 0.5 | 45.1 | 0.1 | 0.5 | 39.5 | 0.3 | 3.3 | 29.5 |

As expected, in oxygen-deficient wastewater environments, concentrations of nitrite and nitrate in the influent water were consistently low (typically below 1 ppm). Under anaerobic conditions, ammonium is not oxidized to nitrite or nitrate because of a lack of oxygen, and any nitrate present tends to be reduced (NO or $N_2O$ and even $N_2$) via denitrification. Additionally, nitrite being chemically unstable tends to be quickly reduced, which explains its low presence across all sampling points.

From influent to settled stage, there is a slight reduction in all forms of nitrogen levels, which reflect the limited removal capacity of the pretreatment and primary treatment. Although nitrification does not occur in this anaerobic stage, ammonium may be partially removed via cellular assimilation by heterotrophic microorganisms present in water.

Another observation from these results is the inverse relationship between influent flowrate and ammonium concentrations. During March and April, increased rainfall causes higher flowrates (March and April with 29192 and 29644 $m^3$/day, respectively), which coincided with lower concentrations at the influent of $NH_4$-N, decreasing from 48.8 in February to 45.1 ppm in March, suggesting dilution effects.

*Table 12* summarizes the calculated removal efficiencies for ammonium and total nitrogen obtained over the monitored period.

*Table 12. $NH_4$-N and TN Removal efficiencies from February-25 to April-25 in Tarragona's WWTP*

| | **Removal Efficiency (%) TN** | **Removal Efficiency (%) NH4-N** |
|---|---|---|
| **February-25** | 27.2 | 19.3 |
| **March-25** | 39.6 | 60.1 |
| **April-25** | 31.6 | 36.1 |

A substantial increase in ammonium and total nitrogen removal was detected in March compared to February despite the relatively stable average temperatures (17.4ºC and 17.8ºC). This difference can be related to the sudden flowrate variation (25749 $m^3$/day to

29192 m$^3$/day) and the subsequent dilution resulting in a raise in the removal efficiencies. However, in April although temperature rose further and flowrates showed minimal variation, both $NH_4$-N and TN removal efficiencies dropped. This is attributed to the temporary shutdown of one of the air blowers in the BR, resulting in less available dissolved oxygen for the oxidation of ammonium.

This effect is further illustrated in *Figure 9*, which displays $NH_4$-N concentration levels in the BRs and the effluent water from April 7$^{th}$ to 11$^{th}$ —corresponding with the blower shutdown period.

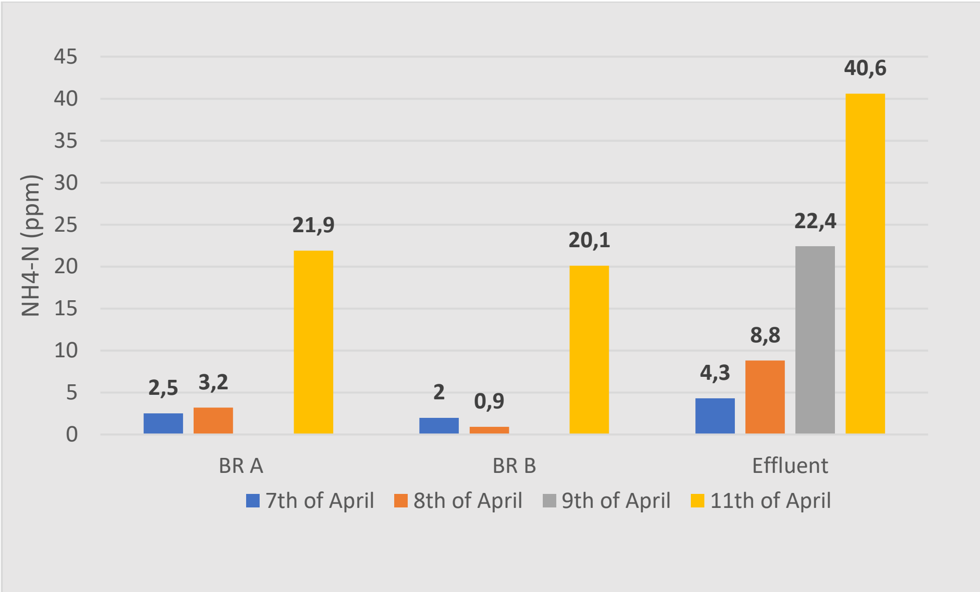


*Figure 9. $NH_4$-N levels before and after air blower shutdown.*

The figure shows a sharp rise in the effluent ammonium levels when the air blower was off (from 8$^{th}$ to 13$^{th}$ April), confirming that oxygen availability was the limiting factor in nitrification. On April 8$^{th}$, the shut off was performed, causing the sudden rise in effluent levels which continuously increase until April 11$^{th}$. Although the flowrate entering the plant was higher than usual because of continuous rainfalls, the oxygen deficit was the dominant factor affecting ammonium oxidation.

Additional insights are provided in *Table 13*, which compares nitrogen species concentrations in both biological reactors.

*Table 13. Monthly levels of nitrate, nitrate and ammonium in Tarragona's BRs*

| BR A (ppm) | | | BR B (ppm) | | | |
|---|---|---|---|---|---|---|
| $NO_2$-N | $NO_3$-N | $NH_4$-N | $NO_2$-N | $NO_3$-N | $NH_4$-N | |
| 0.4 | 1.7 | 21.6 | 0.7 | 2.0 | 20.3 | **February-25** |
| 0.8 | 2.2 | 7.0 | 1.1 | 3.8 | 5.8 | **March-25** |
| 0.4 | 2.2 | 17.3 | 0.7 | 4.3 | 14.9 | **April-25** |

The overall increase of $NO_2$-N and $NO_3$-N from influent to effluent is because of the nitrification-denitrification process occurring in the biological reactors. In the BR, where oxygen is present, ammonium is oxidized to nitrite and nitrate consequently making effluent levels higher than the ones present in the influent.

Throughout the monitored period, BR B consistently exhibited lower ammonium levels and higher nitrite and nitrate concentrations than BR A. This indicates better nitrification performance for BR B, likely due to higher oxygen availability in the reactor, caused by the higher volume entrance of water in the reactor B than in reactor A. Although the two reactors are designed to operate identically, field observations suggest that BR B may receive a slightly higher volume and consequently there is a continuous higher concentration of dissolved and this enhances oxygen transfer promoting a more effective ammonium oxidation.

To further investigate the influence of temperature on the nitrification-denitrification (NDN) process, historical data from July and August 2023 were reviewed (see Annex 1). These months showed significantly higher temperatures (27-29ºC), offering insight into seasonal extremes. Although the dataset is limited, it suggests that at higher temperatures (27–29 °C), microorganisms rapidly assimilate ammonium, causing a marked rise in N-$NO_3$ compared to the monitoring period. Regarding N-$NO_2$, it remains similar to monitoring values because it quickly converts to nitrate. On July $24^{th}$, $25^{th}$ and $26^{th}$, N-$NO_3$ was high overnight (no blower stops), accumulating nitrification. After the first blower stop at 08:55, N-$NO_3$ dropped by 90 %, showing how oxygen removal shifts the process from nitrification to denitrification. On August 2, the first N-$NO_3$ reading was lower due to an additional blower stop from 04:00 to 04:50 that night.

Finally, *Table 14* summarizes TN average monthly concentrations across the sampling points.

*Table 14. TN concentrations at different sampling points from February-25 to April-25.*

| | TN (ppm) | | |
|---|---|---|---|
| | **Influent water** | **Settled water** | **Effluent water** |
| **February-25** | 60.3 | 65.5 | 43.7 |
| **March-25** | 52.1 | 58.3 | 31.2 |
| **April-25** | 56.1 | 60.1 | 38.4 |

The reported data revealed the increase of approximately 5 ppm from the influent water to settled water, despite reductions in nitrite, nitrate and ammonium levels. This likely reflects biological $N_2$ fixation in the primary clarifiers, where low oxygen, low inorganic N, and available carbohydrates favor fixation into organic N[24]. In contrast, aeration tanks and secondary clarifiers have high levels of dissolved oxygen, preventing $N_2$ fixation, so TN does not increase from settled to effluent.

For further discussion, *Table 15* summarizes TN concentrations in BR A and BR B over the monitoring period in Tarragona's WWTP.

*Table 15. TN concentrations in BR A and BR B in Tarragona's WWTP from February-25 to April-25*

| | TN (ppm) | |
|---|---|---|
| | **BR A** | **BR B** |
| **February-25** | 27.6 | 27.5 |
| **March-25** | 23.0 | 22.7 |
| **April-25** | 25.1 | 21.6 |

As previously discussed, since BR B has more nitrification capacity than BR A and moreover BR B gets more water volume, the TN concentrations were consistently lower in BR B. In February, total nitrogen concentrations were nearly identical between both reactors, which is consistent with the similar concentrations observed in nitrate during this month (1.7 ppm and 2.0 ppm). During March, having the lowest values is attributed to the heavy rainfall occurred.

In summary, nitrogen removal efficiency was influenced by different operational variables. While temperature showed a moderate effect, influent flowrate and oxygen availability played more decisive roles.

### 5.2.2 Phosphorus

This section is focused in phosphorus removal, especially in the form of orthophosphate ($P\text{-}PO_4$) and total phosphate (TP). The primary focus was on understanding the behaviors of these phosphorus forms across treatment stages, evaluating the operational conditions, and identifying key correlations between phosphorus content and other parameters such as seasonal temperature.

*Table 16* summarizes the monthly concentrations of orthophosphate in influent, settled and effluent water, along with the calculated removal efficiencies.

*Table 16. Monthly orthophosphate concentrations at different sampling points and removal efficiencies.*

| | | $P\text{-}PO_4$ (ppm) | | |
|---|---|---|---|---|
| | **Removal Efficiency (%)** | **Influent water** | **Settled water** | **Effluent water** |
| **February-25** | 92.1 | 4.4 | 4.1 | 0.3 |
| **March-25** | 97.6 | 4.1 | 3.7 | 0.1 |
| **April-25** | 68.0 | 5.1 | 3.8 | 1.6 |

From the reported data it can be observed that the average concentration of $P\text{-}PO_4$ for influent water ranged from 4.1 ppm to 5.1 ppm. In March, where the sudden variation in the flowrate occurs (from 25749 $m^3$/day to 29192 $m^3$/day), a minor decrease was observed, suggesting dilution effects with rainfall. A more significant variation takes place in April, when effluent concentration is 1.6 ppm despite stable flowrates and higher temperatures. This anomaly aligns with the reactivation of the flotation thickener on April $10^{th}$ after being inactive for operational purposes. This event temporarily reduced phosphorous removal efficiency to 68%. Stability was restored within four days.

*Figure 10* illustrates this operational impact, highlighting the increase in the phosphorus concentrations until the system reached again the stability.

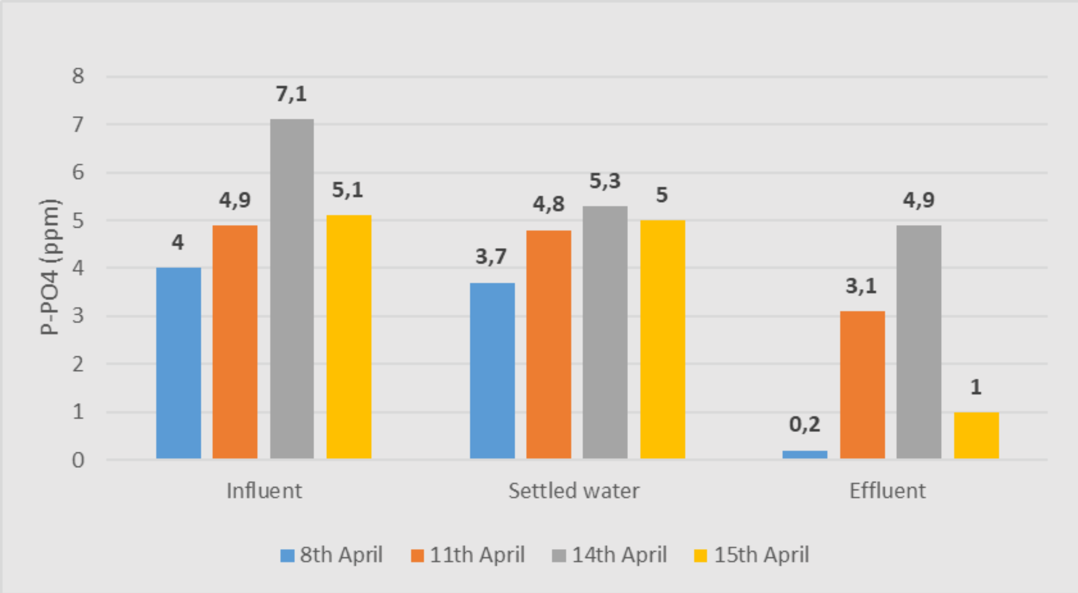


*Figure 10. P-$PO_4$ concentrations before and after flotation thickener return to being active.*

As shown in the graph, both influent and effluent phosphorus levels increased significantly due to internal recirculation from the sludge thickener. On a positive note, the system stabilized within a few days, and effluent phosphorus concentrations returned to their usual values.

To better understand phosphorus dynamics within the biological reactors, further analysis was carried out focusing on orthophosphate and total phosphorus concentrations during the monitoring period. In both BRs, orthophosphate concentrations obtained in February were below 0.015 ppm, suggesting a high capacity for biological phosphorus assimilation by PAOs. This supports additional monitoring of TP in the BRs during March and April.

Since PAOs assimilate phosphorus intracellularly, so a higher SS concentration generally implies greater phosphorus uptake. In other words, phosphorus is not eliminated but transferred from water to sludge[24]. For further discussion, *Table 20* presents TP and SS data for both reactors.

*Table 20. TP and SS concentrations in BR A and BR B.*

| | TP (ppm) | | SS (ppm) | |
|---|---|---|---|---|
| **Date** | **BR A** | **BR B** | **BR A** | **BR B** |
| 11th March | 132.0 | 121.0 | 2933 | 2865 |
| 12th March | 140.0 | 130.0 | 2829 | 2677 |
| 17th March | 150.0 | 153.0 | 1753 | 1871 |
| 7th April | 121.0 | 142.0 | 4740 | 5506 |
| 8th April | 138.8 | 172.5 | 5245 | 5160 |
| 11th April | 159.3 | 111.0 | 4186 | 3815 |
| 14th April | 122.8 | 102.5 | 4208 | 3731 |
| 15th April | 154.8 | 114.0 | 4494 | 4322 |
| 22nd April | 148.5 | 138.0 | 4800 | 4515 |
| 23rd April | 138.3 | 138.3 | 4833 | 4183 |
| 25th April | 170.3 | 156.5 | 4503 | 4745 |
| 30th April | 181.3 | 261.5 | 5156 | 5331 |

The results show a clear trend: total phosphorus (PT) increases with SS levels in the BRs, confirming that phosphorus is removed biologically via PAOs from water and transferred to sludge[25]. Two isolated dates (April 8th and 25th) deviated from the trend. These discrepancies may be related to temporary variations in anaerobic performance, temperature, or dissolved oxygen — which were not monitored during this period.

To confirm the alignment between TP and P-PO4, TP was also measured in influent and effluent water samples throughout April. *Table 21* provides TP concentrations obtained along with the calculated removal efficiency.

*Table 21. TP results obtained in Tarragona's WWTP on influent water and effluent water with removal efficiencies.*

| | | TP (ppm) | |
|---|---|---|---|
| **Date** | **Removal TP Efficiency (%)** | **Influent water** | **Effluent water** |
| **7th April** | 84.1 | 8.2 | 1.3 |
| **11th April** | 10.5 | 7.0 | 6.3 |
| **14th April** | 51.5 | 7.4 | 3.6 |
| **15th April** | 76.3 | 7.1 | 1.7 |
| **22nd April** | 79.7 | 8.0 | 1.6 |
| **23rd April** | 72.1 | 8.3 | 2.3 |
| **24th April** | 79.9 | 8.1 | 1.6 |
| **25th April** | 92.3 | 7.7 | 0.6 |
| **28th April** | 94.1 | 8.3 | 0.5 |
| **30th April** | 79.5 | 6.7 | 1.4 |

The data show that phosphorus removal was generally effective, with removal efficiencies higher than 75%. There were exceptions as on April 11th and 14th (with values of 10.5% and 51.5%, respectively), immediately following the thickener reactivation. These values were associated with high phosphorus concentrations in the effluent and also with elevated turbidity (~15 NTU when the typical is 3–5 NTU), suggesting partial sludge washout.

In summary, phosphorus removal in Tarragona's WWTP is driven primarily by biological assimilation by PAOs, which transfer phosphorus from water to sludge, indicating the proportional relationship between SS and PT. The occasional increases of phosphorus levels in effluent were attributed to operational changes, particularly the reactivation of specific process units as flotation thickener.

## 5.3 Correlation between BRs Temperature and Removal Efficiencies

This section evaluates whether the temperature of the biological reactors has an effect on the removal efficiencies of the nutrients. To this end, scatter plots (x,y) were generated using MATLAB, plotting removal efficiencies of ammonium ($NH_4$-N), total nitrogen (TN), total phosphorus (TP), and orthophosphate (P-$PO_4$) against the BRs temperature values recorded from February 25 to April 25.

This graphical analysis enabled a visual inspection of potential trends or correlations between temperature and treatment performance and facilitated the Pearson's coefficient to empirically support trends or patterns observed in the graphical representations.

### 5.3.1 Nitrogen

*Figure 11* presents the scatter plot obtained by MATLAB showing the relationship between the BRs temperature and the removal efficiency of ammonium (N-$NH_4$) on different operational days from February-25 to April-25.

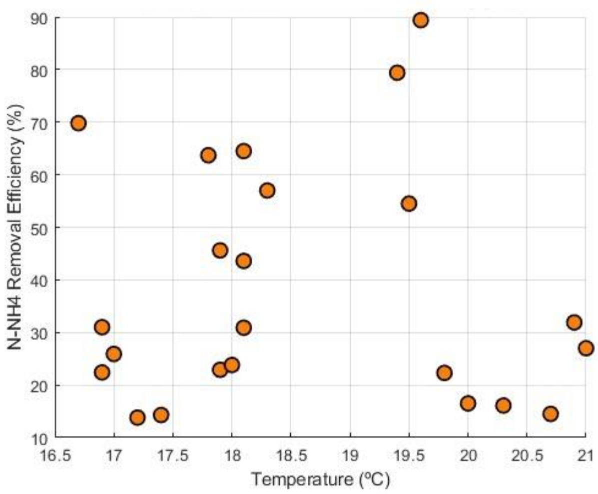


*Figure 11. Temperature vs $NH_4$-N Removal Efficiency.*

Although the obtained data shown in the figure presents some variability, a general moderate trend can be observed, suggesting that higher temperatures tend to correspond to improved ammonium removal efficiencies. However, this correlation is not completely linear, indicating that other operational factors also play a significant role. For instance, in the higher temperature range (approximately 19.5 to 21 °C), there is a notable drop in removal efficiency, which contrasts with the expected trend. This behavior can be explained by a temporary shutdown of one of the reactor's aeration blowers (April 7$^{th}$ to April 11$^{th}$), which limited oxygen availability and negatively affected the nitrification process.

Similarly, at 17.2 °C and 17.4 °C, unexpectedly lower efficiencies were recorded. No changes in flow rate or operational conditions were identified for these days, so these are considered isolated events that remain unexplained based on available data. Conversely, the high removal efficiency of around 70 % at 16.7 °C appears unusually elevated for that temperature, likely caused by a sudden inflow increase following to rainfall (30545 $m^3$/day), artificially increasing the removal efficiency for that day.

To empirically assess the potential correlation between temperature and ammonium nitrogen removal efficiency, the Pearson correlation coefficient (r) was calculated. The last six data points, corresponding to the 20–21 °C range, were excluded due to the justified operational shutdown of the air blower that affected performance. The resulting coefficient of 0.61 suggests a moderate positive correlation between the BR temperature and $NH_4$-N removal efficiency.

A similar analysis was performed for TN removal as shown in *Figure 12*.

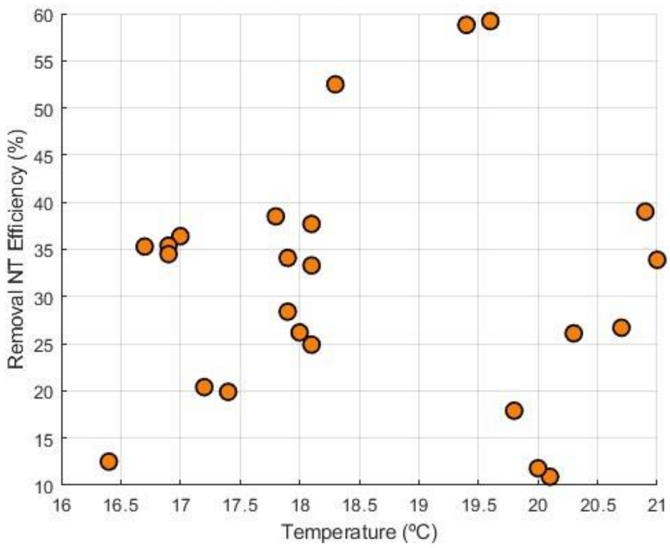


*Figure 12. BRs Temperature vs TN Removal Efficiency*

The scatter plot suggests that higher temperatures generally correlate with improved TN removal. However, some operational issues also influence performance. At 20–21 °C, removal dropped sharply due to a blower shutdown—these points were excluded from the correlation. Two points at 17.2 °C and 17.4 °C also showed unexpectedly low TN removal, likely due to unmonitored variations.

The cluster of data points between 16.5ºC to 17.0ºC, corresponds to days with higher than-average influent TN concentrations (around 65 ppm TN), which resulted in higher-than-expected removal. Conversely, the three highest efficiency values coincided with slightly lower effluent TN, increasing calculated removal.

After excluding blower-related data, the Pearson correlation coefficient between temperature and TN removal was r = 0.71, indicating a moderate to strong positive correlation between BR temperature and TN removal efficiency.

### 5.3.2 Phosphorus

To explore the relationship between BR temperature and the removal efficiency for P-$PO_4$ scatter plots were also generated by MATLAB. *Figure 13* shows P-$PO_4$ removal efficiency against BR temperature.

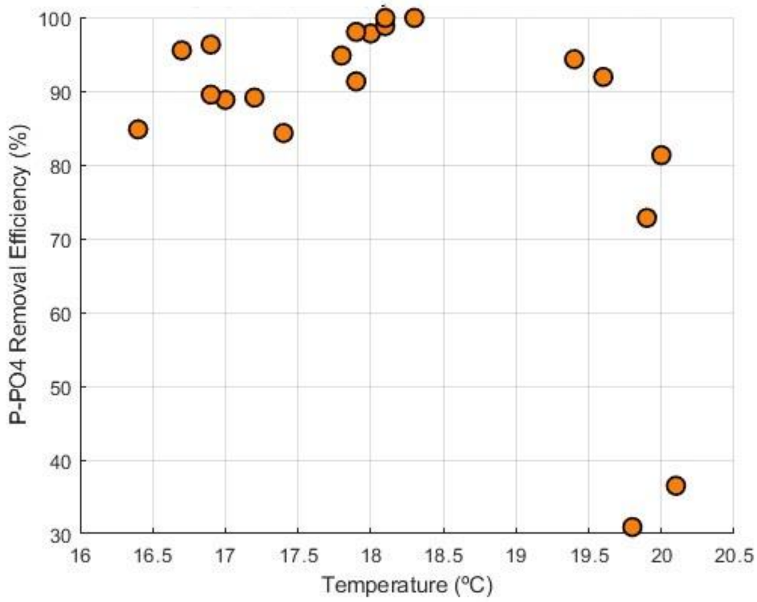


*Figure 13. BRs Temperature vs P-$PO_4$ Removal Efficiency*

Most phosphate removal efficiencies exceed 90 %, indicating robust biological phosphorus assimilation by PAOs. However, there were isolated deviations from this pattern. At 16.4 °C and 17.4 °C efficiencies decreased slightly to around 80-85%, corresponding to slightly higher effluent P concentrations (~0.7 mg/L) compared to the typical values (0.3 ppm-0.5 ppm).

The lowest values (below 40%) occurred during flotation thickener reactivation when the temperature was between 19.5 °C and 20 °C caused by the internal sludge returns previously mentioned.

After excluding the four thickener-affected data points, the Pearson coefficient was calculated with a value of r = 0.40, indicating a weak association between BR temperature and orthophosphate removal.

A parallel analysis was performed with TP, monitored only in April due to reagent limitations. *Figure 14* plots the removal TP efficiencies against BRs temperature to visualize any possible correlation between these two parameters.

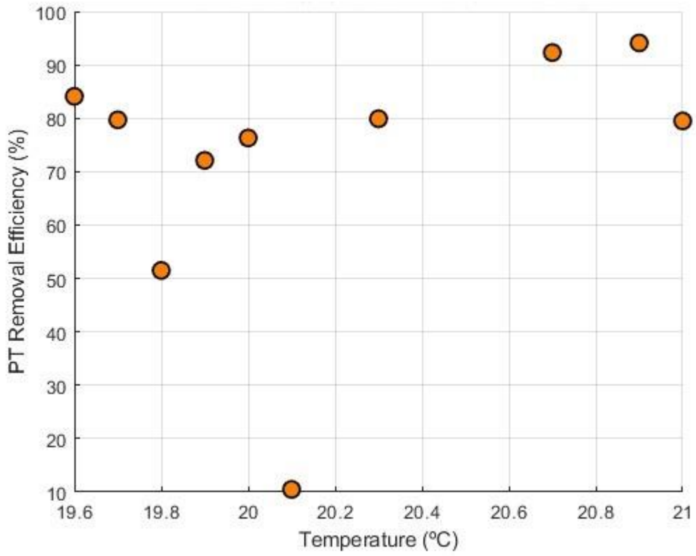


*Figure 14. BRs Temperature vs TP Removal Efficiency*

TP removal efficiencies follow a similar pattern to P-$PO_4$, with generally satisfactory results—especially considering there are no regulatory phosphorus limits at the Tarragona WWTP.

Two low-efficiency data points (10.9% at 20.1 °C and 51.5% at 19.8 °C) from April 11th and 14th align with P-$PO_4$ deviations linked to flotation thickener reactivation. Excluding these, the correlation coefficient is r = 0.53, suggesting a moderate relationship between BR temperature and TP removal. However, the limited number of measurements makes it difficult to draw strong conclusions.

In summary, the correlation indicates that nitrogen removal has a stronger correlation with BR's temperature while for phosphorus, the obtained value is not strong enough to confirm a clear temperature influence. The findings confirm that temperature is an important contributing factor but there are other factors such as operational parameters, dissolved oxygen, influent load, hydraulic overloads that also play a fundamental role in the nutrient removal. Therefore, when assessing or optimizing nutrient removal, these parameters must be considered alongside with temperature to obtain a more accurate picture of the system behaviour.

# 6.CONCLUSIONS

The main conclusions derived from this study aimed to evaluate the nutrient removal behaviour and system stability at the Tarragona's WWTP are as follows:

- This study has successfully established expected concentration ranges for key nutrients (nitrogen and phosphorus) across different sampling point within the Tarragona wastewater treatment plant. These values are greatly valuable and serve as a useful reference for future studies.
- Key parameters (BOD5, COD and SS) have shown high and consistent removal efficiencies throughout the monitored period. Control charts confirm stable operation, often exceeding design values despite seasonal and hydraulic variations.
- Occasional operational incidents, such as temporary air blower shut down or the reactivation of the flotation thickener, affected more negatively to the removal efficiency than seasonal temperature variations in the biological reactors. These events cause temporary deviations but the system soon returned to normal and regained stability.
- A moderate positive correlation was observed between BR temperature and nitrogen removal efficiency (r= 0.61 for $NH_4$-N and r = 0.71 for TN). In contrast, the correlation with phosphorus removal (r = 0.40 for $PO_4$-P and r = 0.53 for TP) was weaker, due to other operational factors.
- Phosphorus removal has been consistent and high, indicating that the plant is well-prepared for stricter regulations. However, nitrogen removal may require improvements if regulatory requirements become more stringent.
- This study has primarily focused on the relationship between temperature and nutrient removal efficiencies. Further analysis should include additional variables such as dissolved oxygen or influent load, benefiting from considering the combined influence of other operational and environmental parameters. It is recommended to continue the study with longer time series and a multivariate analysis to optimize the plant operation under varying conditions.

# 7.ANNEX

*Annex 1. Concentrations of nitrogen in the form of nitrite and nitrate in BR A (Historical data from Summer 2023)*

| | 24th July 2023 (28,3ºC) | | 25th July 2023 (27,6ºC) | | 26th July 2023 (26,9ºC) | | 2nd August 2023 (27,9ºC) | |
|---|---|---|---|---|---|---|---|---|
| | **$N\text{-}NO_2$ (ppm)** | **$N\text{-}NO_3$ (ppm)** | **$N\text{-}NO_2$ (ppm)** | **$N\text{-}NO_3$ (ppm)** | **$N\text{-}NO_2$ (ppm)** | **$N\text{-}NO_3$ (ppm)** | **$N\text{-}NO_2$ (ppm)** | **$N\text{-}NO_3$ (ppm)** |
| **8:00 (before)** | 0,278 | 14,8 | 0,362 | 17,1 | 0,676 | 9 | - | 2,3 |
| **8:55 (after)** | - | 1,4 | - | 1,7 | - | 0,8 | - | 1,3 |
| **10:00 (before)** | - | 8,4 | - | 8,1 | - | 3,9 | - | 3,9 |
| **10:55 (after)** | - | 2,4 | - | 2,5 | 0,020 | 1,1 | - | 1,9 |
| **13:00 (before)** | - | 6,8 | - | 2,6 | - | - | - | - |
| **13:55 (after)** | 0,012 | <1 | 0,013 | 1,2 | - | - | - | - |

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