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Review

Nutrient Removal and Recovery from Municipal Wastewater

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Abstract: With the ongoing amendment of the EU legislation on the treatment of urban wastewater, stricter requirements for the removal of pollutants are expected, which calls for the need for innovative wastewater treatment technologies. Biological systems are still the first choice. A survey of typical bioreactors applied in wastewater treatment is presented. The wastewater treatment objective, biochemical environment, and microbial growth are selected as the main criteria for the classification of these bioreactors. Hydraulic and kinetic aspects are considered, along with the advantages and drawbacks of these bioreactors regarding the selection of the appropriate type of reactor; as well, details regarding the operation of reactors are mentioned. The aim of this paper is to provide operators and designers with a brief overview of the selected traditional and advanced processes, reactors, and technologies for nutrient removal from municipal wastewater. The possibilities and limitations in complying with more strict effluent standards are also discussed. Methods of nutrient recovery are added value. From the evaluation of the published papers, we determine that the currently applied traditional methods for nutrient removal have the potential to also convey the expected stricter limits.

Keywords: municipal wastewater; nutrient; removal; recovery; secondary; tertiary; treatment

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Citation: Derco, J.; Žgajnar Gotvajn, A.; Guľašová, P.; Kassai, A.; Šoltýsová, N. Nutrient Removal and Recovery from Municipal Wastewater. *Processes* **2024**, *12*, 894. https://doi.org/10.3390/pr12050894

Academic Editors: Andrea Petrella and Avelino Núñez-Delgado

Received: 20 February 2024
Revised: 26 March 2024
Accepted: 26 April 2024
Published: 28 April 2024

1. Introduction

Wastewater (WW) is essentially the water supply of a community from a variety of uses. From the point of origin, wastewater may be characterized as a combination of the liquid and solid waste removed mainly from residences, social and industrial institutions, and similar facilities together with groundwater and stormwater, depending on the type of sewer system. Domestic wastewater means wastewater from residential settlements and services that originates from the human metabolism and from household activities [1]. Municipal wastewater is water delivered to communities after various uses originating from domestic, industrial, commercial, and institutional sources within a given human settlement or community [2]. Urban wastewater includes both municipal wastewater and urban runoff. The accumulation of untreated wastewater can lead to the production of malodorous gases due to the decomposition of organic impurities. In addition, untreated wastewater usually contains numerous pathogenic or disease-causing microorganisms. It usually contains nutrients, which stimulate the growth of algae and other aquatic plants and consequently accelerate eutrophication. A high content of organic and ammonium impurities can lead to the excessive consumption of dissolved oxygen and undesirable changes in the aquatic life of the receiving water. Wastewater can also contain toxic compounds. It is therefore a necessity to treat wastewater before discharging it into the receiving water. Nitrogen and phosphorous are inevitable for the growth and vitality of living beings. However, the discharge of nutrient-rich wastewater into aquatic ecosystems leads to eutrophication. Thus, the effective removal of nutrients N and P from WW is a way to meeting strict nutrient discharge standards [2]. In addition, nutrient recovery from WW is equally important for realizing a circular economy by preventing the depletion of limited resources.

On 26 October 2022, the European Commission published a proposal [3] for the revision of the Council Directive from 21 May 1991 on the treatment of municipal wastewater: "Urban Wastewater Treatment Directive—UWWTD". Although the presented proposal represents material that is still subject to debate and changes, it can be considered as the framework and basic theses that this proposal contains. The proposed directive [3] emphasizes not only the protection of the environment but also the protection of people's health, the reduction of greenhouse gas emissions, better access to sanitation and wastewater treatment, an improvement in management and transparency, and an improvement in the information sources on public health. The original directive [2] went through several evaluations during its validity. The European Commission published its latest assessment (2019 REFIT EVALUATION) [3] in December 2019. This assessment shows significant reductions in pollutants in discharged waste in terms of the BOD_5, total nitrogen, and total phosphorus, and thus, a significant impact on the quality of lakes, rivers, and seas in the EU are visible and tangible. However, the evaluation also pointed out three complex and ambitious challenges:

- Remaining pollution:

A decrease in the pollution load from individual sources in the EU is possible by reducing pollution discharged from overflows of relief chambers (Stormwater Overflows, SWOs), from agglomerations below 2000 EO or inadequate individual treatment systems.

- Insufficient alignment of the directive with the objectives of the EU policy around the EU Green Deal:

Decreasing the total energy consumption and all greenhouse gas emissions in the whole EU is possible by improving the treatment processes, making better use of sewage sludge, increasing the energy efficiency, and achieving a higher rate of use of renewable resource technologies. It is necessary to better integrate this area into the circular economy to improve sludge management and the reuse of water and valuable resources.

- Insufficient and uneven level of management:

The evaluation report also pointed out the different levels of operation of sewage networks and wastewater treatment plants between individual EU member countries. The existing monitoring and reporting do not use all the possibilities of digitization. The results of the public consultation resulted in a request to address the area of micropollutants in wastewater and to better apply the "polluter pays" principle by introducing and expanding producer responsibility. With the ongoing amendment of EU legislation on urban wastewater treatment [3], stricter requirements for pollutant removal are expected, driving the need for innovative environmental technologies. Diverse pollutants in urban wastewater, including macronutrients and micropollutants, require advanced treatment technologies that integrate biological, physical, and chemical processes [4].

This review article is focused on the removal and recovery of nutrients N and P from municipal wastewater. It provides the descriptions of processes, reactors, and technologies as well as new challenges in the context of the proposed directive [4]. The aim of this paper is to provide operators and designers with a brief overview of the selected traditional and advanced processes, reactors, and technologies for nutrient removal from municipal wastewater. Another goal is to discuss the possibilities and limitations in complying with more strict effluent standards.

Most wastewater treatment plants (WWTPs) are equipped with mechanical pretreatment, where floating substances (rakes) and settling inorganic solids are physically removed by sedimentation (sand traps). In the primary treatment, undissolved organic substances are separated in sedimentation tanks [5]. In the secondary treatment, the remains of organic substances are removed using biological processes, while macronutrients (N and P) are also partially removed by their assimilation into the new cell mass. Thus, the main function of conventional WWTPs is to remove organic pollution. However, even after a typical secondary treatment, some dissolved and suspended substances remain in WW along with the main parts of N and P [6–8]. From the point of view of macronutrients, they therefore represent a dominant point source of nutrient pollution in the aquatic ecosystem with N and P [8]. A tertiary or advanced treatment system is required for their desired removal. Due to the dominant applications of biological processes using activated sludge in sewage treatment practices [9,10], these processes also began being used in tertiary technologies for the removal of macronutrients. Wastewater treatment facilities are subject to real conditions that impact the performance and variability of effluent concentrations. These conditions include seasonal challenges such as lower temperatures, wet weather high-flow events, changes in influent characteristics, unavoidable imperfections that are present in every design or operation, mechanical problems, and impacts of toxic discharges into the sewer collection system that impact plant process performance, among others.

It is necessary to state that the high operating and investment costs associated with the implementation of these processes and technologies require significant financial requirements, and thus, this introduces limitations to their wide use in technological practices [5]. It turns out that similar limitations also apply to physical methods. Membrane applications are also costly. They are not very effective, as they only remove approximately 10% of the total macronutrients. Other problems include membrane fouling [11,12]. Chemicals are also expensive. Handling and storing them is hazardous. Their applications lead to higher sludge formation, which increases maintenance and operation costs [13].

2. Nutrient Removal

It is well known that biological processes are widely used for the removal of organics, nitrogen, and phosphorus from wastewater. One reason is the possibility to achieve a high elimination efficiency of these pollutants from wastewater. In addition to this, there are relatively low capital and operational costs in comparison to physicochemical or chemical processes [14]. Biological processes of wastewater treatment are also environmentally friendly. They represent the intensification of the processes that occur in nature, for example, the well-known process of self-purification in rivers or the rotting of organics (break down/decay) in the absence of air. The intensification of natural biological processes is carried out in bioreactors, resulting in the removal of organic, nitrogen, and phosphorus substances, which increases the rate of the treatment process and consequently minimizes the required reactor volumes, capital, and operational costs [15,16].

The draft of the new directive [3] establishes the conditions for ensuring the tertiary treatment of municipal wastewater (nitrogen and phosphorus). The concept of sensitive areas is abolished, and the new Member States will have the obligation to define areas sensitive to eutrophication. The main change is the fact that the requirements for nitrogen and phosphorus removal are not determined by the size of the agglomeration, but directly by the size of the load on individual WWTPs. In the case of WWTPs with a size of over 100,000 PE, a tertiary treatment will be mandatory. For WWTPs with a size of over 10,000 PE, they will be mandatory if their treated wastewater is discharged into an area defined as sensitive to eutrophication. The original concentrations for total nitrogen of 15 mg/L (for 10,000–100,000 PE) or 10 mg/L (at more than 100,000 PE) are reduced to 6 mg/L. The corresponding original minimum reduction values for the total nitrogen are 80%, or 70–80%, which will increase to 90, or 85%, respectively. The original values for the total phosphorus of 2 mg/L (for 10,000–100,000 PE) or 1 mg/L (at more than 100,000 PE) are reduced to 0.5 mg/L. The corresponding original minimum reduction value for the total phosphorus of 80% is increased to 90%. Depending on the local situation, either or both parameters may be used. One of the main topics of the transposition of the future directive [3] will therefore be the definition of areas sensitive to eutrophication.

Practical applications show that the conventional activated sludge process with a gradual flow can be easily intensified to a system with biological phosphorus removal by creating an anaerobic zone at the beginning of the activation tank. This reconstruction requires the installation of a baffle to prevent back mixing, the shutdown of aeration, and the installation of an anaerobic mixing zone [17]. This system is highly efficient in terms of phosphorus removal if nitrification does not occur under oxic conditions. On the other hand, this system can also be modified for the purpose of nitrogen removal, namely by installing additional baffles, mixers, and including the appropriate recirculation of nitrates.

The development of requirements for the quality of treated water, requirements for cost reduction, and the development of processes and technologies for treating WW are also shifting significantly towards hybrid processes that use combined forms of biomass [4].

Biological nutrient removal (BNR) is a term used to describe the application of microbes to take up nitrogen and phosphorus during WW treatment [18]. As the requirements for the quality of treated WW increase, so does the complexity of their treatment technologies. Individual biological processes for the removal of organic pollution, N, and P require the provision of different conditions in terms of the presence of oxygen (oxic, anoxic, or anaerobic conditions) as well as the required form of carbon (inorganic or organic) [15,16]. No less important are the differences in the values of specific growth rates of dominant microorganisms for individual processes and their sensitivity to the presence of various pollution components, temperature changes, technological parameters, etc. These factors must be considered when designing the processes and technologies for treating WW as well as during their actual operation. On the other hand, the removal of nutrients from WW is also accompanied by higher costs [19].

2.1. Nitrogen Removal Processes

Nitrogen is one of the basic elements of cell mass. However, its excessive release into water bodies can lead to the acceleration of eutrophication in the receiving water. Nitrogen removal from wastewater can be carried out using physicochemical, chemical, bioelectrochemical, and biological methods. The authors of [20] dealt with the operating conditions, advantages, and disadvantages of used processes and influencing factors. Ammonia stripping by air is the most common pretreatment process. However, it requires large spaces and is time consuming. Its disadvantage is also the formation of calcium carbonate deposits. Chemical processes showed the least efficiency around 20–30%, which is dependent on the pH of the process. Biological treatment processes (nitrification, denitrification, and anammox) are among the most effective (in terms of the achieved efficiency (>95%), costs, and environmental origin) for the removal of nitrogenous pollutants from municipal WW. Due to the complexity of wastewater, a combination of different methods using the advantages of individual systems is recommended for the efficient and economical treatment of WW in accordance with the characteristics of WW and the quality requirements.

Ammonium nitrogen can be transferred to nitrite or nitrate in an aerobic environment. Inorganic carbon, i.e., carbon dioxide, is utilized by chemoautotrophic nitrification bacteria (*Nitrosomonas* and *Nitrobacter*) in the synthesis of new microorganisms. The energy-yielding two-step oxidation of ammonia to nitrate is generally represented as follows [17]:

Nitrosomonas

$$2NH_4^+ + 3O_2 \rightarrow 2NO_2^- + 4H^+ + 2H_2O \tag{1}$$

Nitrobacter

$$2NO_2^- + O_2 \rightarrow 2NO_3^- \tag{2}$$

The total reaction is expressed as

$$NH_4^+ + 2O_2 \rightarrow NO_3^- + 2H^+ + H_2O \tag{3}$$

The removal of the products of nitrification can be accomplished in anoxic conditions, i.e., the level of dissolved oxygen is maintained near zero (less than 0.5 mg/L). The reduction of nitrite and nitrate to nitrite, to nitric oxide, to nitrous oxide, and finally to nitrogen occurs. In anoxic conditions, i.e., where the level of dissolved oxygen is maintained near zero (less than 0.5 mg/L), the reduction of nitrite and nitrate to nitric oxide and then to nitrous oxide/nitrogen occurs according to Equation (4):

$$NO_3^- \rightarrow NO_2^- \rightarrow NO \rightarrow N_2O \rightarrow N_2 \quad (4)$$

Nitrate nitrogen is utilized as the terminal electron acceptor, releasing nitrogen gas as the product in the process of denitrification. An organic carbon source is necessary in the process of denitrification.

By combining shortened nitrification, so-called nitritation, significant savings in both the investment and operating costs can be achieved with the anammox process. Nitrifying bacteria (AOB, ammonium oxidizing bacteria) oxidize 57% of the input NH_4^+ to NO_2^- according to Equation (5), which saves approx. 50–60% of energy for aeration compared to nitrification. The reason for this is that only part of the ammoniacal nitrogen is oxidized biochemically, while the oxidation is already completed in the nitrite stage. Anammox microorganisms transform the remaining ammoniacal nitrogen and gaseous N_2 according to Equation (6) [21]. As the anammox microorganisms of the process belong, unlike denitrifying bacteria, to chemolithotrophic microorganisms, nitrogen is essentially removed without the consumption of organic substrate. At the same time, up to 80% less excess sludge is produced. The anammox process transforms up to 13% of the input nitrogen into nitrates. In practice, these nitrates are at least partially denitrified by heterotrophic microorganisms. The advantages of technologies based on the anammox process include a high performance (the proposed load can be 0.35–2.3 $kg_N/(m^3/d)$ [22], and in some cases, even 10 $kg_N/(m^3/d)$ [23]).

$$NH_4^+ + 1.5\,O_2 \rightarrow NO_2^- + 2H^+ + H_2O \quad (5)$$

$$\begin{aligned} &NH_4^+ + 1.32\,NO_2^- + 0.066\,HCO_3^- + 0.13\,H^+ \\ &\quad \rightarrow 1.02\,N_2 + 0.26\,NO_3^- + +0.066\,CH_2O_{0.5}N_{0.15} + 2.03\,H_2O \end{aligned} \quad (6)$$

Biological nitrogen removal (BNR) is a critical process in WW treatment. The authors of [24] present new microorganisms that can remove nitrogen through new metabolic pathways. These include ammonia oxidizing archaea (AOA), complete ammonia oxidation bacteria (COMAMMOX), anaerobic ammonium oxidation associated with iron reduction bacteria (FEAMMOX), anaerobic ammonium oxidation bacteria (ANAMMOX), and anaerobic methane oxidation denitrifying microorganisms (DAMOs). Compared to current nitrifying or denitrifying bacteria, these new microbial groups have better a physicochemical tolerance and/or lower release of greenhouse gases. They represent promising microbial communities that could provide new technologies for high-performance and energy-saving nitrogen removal from WW.

2.1.1. Single Anoxic System

The development of nitrogen removal technologies gradually progressed from three-sludge to single-sludge systems. In the case of two-sludge systems, it is advantageous to operate the denitrification process in reactors with increased biomass (packed columns, rotating disk reactors, and fluidized bed bioreactors). Systems with separated biocenosis are relatively stable and achieve high nitrogen removal efficiencies. However, their disadvantage is relatively high investment and operating costs.

The choice of nitrogen removal technology is marked not only by the characteristics of wastewater, the required quality of treated water, and investment options but also by the stage of decision making and selection. In the case of the construction of a new sewage treatment plant (WWTP), it is possible to consider all aspects more comprehensively, starting with the requirements for the built-up area, ending with guarantees of quality for the outflow and flexibility in terms of managing individual processes in the event of changes in the amount and composition of wastewater. Currently, single-sludge systems are most often operated. Under single-sludge systems, this refers to systems in which activated sludge is separated from treated wastewater in only one settling tank. These systems make it possible to use organic wastewater pollution as a source of organic carbon for denitrification, while relatively high nitrogen pollution-removal efficiencies are also achieved. The most widespread arrangements of a single-sludge system regarding the removal of nitrogen compounds from wastewater include upstream denitrification and simultaneous denitrification. In the case of some industrial wastewaters, including denitrification appears to be advantageous. These systems make it possible to achieve a relatively high treatment efficiency also when using municipal WW as a source of organic carbon for denitrification. They are considerably flexible in terms of the possibility of controlling the nitrification and denitrification processes. The most used single-sludge system technologies for removing nitrogen compounds from WW are upstream denitrification and simultaneous denitrification. Each system with a single anoxic zone (single anoxic system) should be able to achieve, for sewage WW, average annual output values for total nitrogen (TN) of 8–12 mg/L. [25].

Most of the biological stages of urban WWTPs can be characterized from the point of view of the hydrodynamic regimen as systems with a gradual flow. The investigation of the influence of hydrodynamic conditions on the kinetics of the nitrification process by Chudoba et al. [26] showed that the gradual flow system is more advantageous for the nitrification process compared to the mixing system [26]. Azimi and Horan [27] also presented the advantages of a gradual flow nitrification system under steady state conditions [27]. Similar results were also published by Horan and Azimi [28] based on their investigation of the responses of nitrification systems with different hydrodynamic conditions to a sudden increase in nitrogen pollution [28]. Both systems showed very similar responses for the monitored forms of nitrogen pollution. Thus, in the mixing system, the effect of dilution in favor of the nitrification process was not observed.

Wuhrmann System

BNR processes can occur in a two-step process that begins with the use of autotrophs to oxidize NH_4^+ to NO_3^- (nitrification), followed by the reduction of NO_3^- to N_2 by heterotrophic microorganisms (denitrification). In this context, it should be emphasized that autotrophic bacteria necessarily need dissolved oxygen to oxidize ammonia nitrogen, even if they are able to survive for a long time in an environment without its presence. This ability of theirs is also very important from a practical point of view, as it enables nitrification and denitrification processes to take place in one bioreactor. Nitrifying bacteria are much more sensitive and vulnerable compared to heterotrophic microorganisms. Since they are slow growing, their retention in the system requires higher solid retention time (SRT) values [29]. According to Equations (1)–(3), 4.57 g of oxygen are theoretically consumed for the oxidation of 1 g of ammoniacal nitrogen. After considering the consumption of ammonia nitrogen for the synthesis of nitrifying biomass, the specific consumption of oxygen for oxidation is about 4.3 gO_2/gNH_4-N. A value of 2 mg/L O_2 is considered a safe concentration of dissolved oxygen to ensure the nitrification process. At the same time, it reduced silamentous sludge formation. If there is a drop in the oxygen concentration, the most endangered are the autotrophic bacteria responsible for an efficient nitrification process. Nitrifying bacteria are mostly found inside activated sludge flocks, where the dissolved oxygen concentration values are lower compared to the surrounding liquid. If there is a higher material load (typical for the Wuhrmannn system), the oxygen consumption will increase, and the concentration of dissolved oxygen in the flocs may drop more. Therefore, it is important to monitor and control the supply of oxygen in the oxic tank/zone of the MWWTP. The system was developed by Wuhrmann [30], and in the literature, it is referred to according to its developer or post-denitrification system. In the case of domestic wastewater, the basic disadvantages of this system include the need for an external source of organic carbon as well as the increased cost of the oxygen supply in connection with the oxidation of organic wastewater pollution in the aerobic stage. In this system, it is not possible to eliminate the decrease in the neutralization capacity due to the nitrification process by hydroxyl ions produced in the denitrification stage. For this system, it is also necessary to include post-aeration before the settling tank. Post-denitrification is advantageous mainly from the point of view of the possibility of achieving a higher efficiency in the process but also from the point of view of energy requirement (smaller requirements for recirculation) compared to pre-denitrification. Thus, to achieve the required nitrogen removal efficiency, the supply of an external *bio*carbon source that does not contain ammonia or organic nitrogen, or has a small content, is inevitable. Monteith et al. [31] used, as an organic carbon source for denitrification, 30 different types of industrial wastes and compared the results with denitrification using methanol as an organic carbon source. Among the wastes used were various food (brewery, distillery, wine, and starch), chemical (organic acids, distillation residues, and higher alcohols), and other waste products. In 27 cases, higher denitrification rates were achieved than when using methanol. In addition to liquid industrial waste, it is also possible to use gases, especially methane and natural gas. The basic conditions for all carbon sources for denitrification are their good biological degradability and low or no toxicity to the nitrification process [31].

The complete elimination of oxide forms of nitrogen in the Wuhrmann system is possible, but it is conditional on the availability of suitable organic carbon.

Modified Ludzak–Ettinger System

Ludzack and Ettinger (LE) [32] developed a concurrent system by placing an anoxic zone ahead of the oxic zone. The LE system was improved by Barnard [25,33] by providing additional internal recirculation to return the mixed liquor back from the nitrification to the denitrification zone. Such a system in the literature is referred to as a Modified Ludzack–Ettinger (MLE) or pre-denitrification system. In upstream denitrification in an MLE system (Figure 1), WW first flows through an anoxic zone, in which the organically bound nitrogen is hydrolyzed to ammoniacal nitrogen. The ammonia contained in wastewater is consumed in the anoxic zone only for the synthesis of new biomass. In the second, the oxic zone, ammoniacal nitrogen is oxidized to nitrites and nitrates. Oxidized forms of nitrogen are transported back to the anoxic zone by the internal recirculation (IR) of mixed liquor suspended solids (MLSSs) as well as the external recycling of return sludge (RS). Heterotrophic microorganisms use organic pollution from WW as a source of carbon, while oxidized forms of nitrogen are reduced to N_2O or nitrogen gas. The efficiency of the denitrification process depends on the amount of recirculation. On the other hand, with a higher amount of recirculation, there is a danger that a large amount of oxygen is introduced into the anoxic zone, which inhibits denitrification. It is possible to prevent this effect, e.g., by including a small anoxic zone at the end of the oxic part of the system. Due to the higher specific rates of denitrification compared to nitrification, the volume of the denitrification part is generally smaller than that of the nitrification zone. This volume is also often used to increase the volume of the oxic part of the activation, especially in the winter months, due to a significant decrease in the rate of nitrification compared to the summer period [25]. Another problem is the drop in the pH during nitrification. Each nitrified mg of NH_4-N corresponds to the production of 0.14 mmol of H^+. The recommended MLSS pH value is in the range of 7 to 8. Lowering the pH below 7 can cause a decrease in the rate of nitrification. For this reason, pH monitoring and the possibility of dosing an alkaline agent during the activated sludge process is necessary. The temperature dependence of nitrifying bacteria is also an important factor. It has been common practice until now that effluent concentrations of reduced forms of nitrogen were defined for winter temperatures to avoid the oversizing of the system by forming an unused volume during the longer, warmer period of the year. An alternative is the flexible use of certain volumes/sections by increasing the oxic volume at lower/winter temperatures and increasing the anoxic volume at higher/summer temperatures [29].

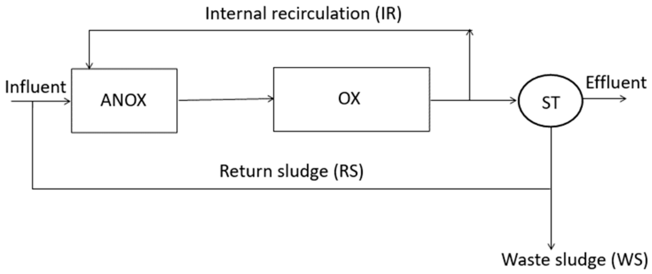

Figure 1. Scheme of Modified Ludzak–Ettinger system (MLE). ANOX—anoxic zone; OX—oxic zone; ST—settling tank. (ST).

If there is a sufficiently low concentration of dissolved oxygen, microorganisms use organic pollution from wastewater as a source of organic carbon, and oxidized forms of nitrogen are reduced to N_2O or nitrogen gas. The activity of denitrification affects the amount of recirculated liquid, R = (RS + IR), according to Equation (7) [27]:

$$E = \frac{R}{R+1} 100\% \tag{7}$$

From Equation (7), following that at a value of the total recirculation ratio R of 4, about 80% of nitrates are removed by denitrification. A further increase in recirculation would be accompanied by a disproportionate increase in the operating costs compared to the achieved effect in terms of the denitrification efficiency. In addition, at higher values of recirculation, a larger amount of oxygen can be introduced into the anoxic zone, which inhibits the denitrification process. It is possible to prevent this effect, e.g., by including a small anoxic zone at the end of the oxic part of the system [25]. Another disadvantage of this arrangement is the decrease in the concentration gradient of the oxic part due to internal recirculation, which can be reflected in the deteriorated sedimentation properties of the sludge. An MLE type of approach can be relatively easily retrofitted into an existing wastewater treatment plant through the installation of baffles, mixers, and an internal recycle capacity. ln a diffused air system, the diffusers would have to be relocated to create an anoxic zone and to ensure that there is adequate air for nitrification in the oxic zone. Some of the diffusers could be left in the anoxic zone to provide mixing if the dissolved oxygen concentrations were kept at a low level. The problem of an insufficient oxygen concentration may also be related to increasing the SRT of the sludge to intensify the nitrification process. Nitrifying bacteria are slow growing. An increase in the age of the sludge is associated with an increased concentration of bacteria, and thus, a higher oxygen consumption. The intensification of the nitrification process by increasing the age of the sludge or biomass concentration in the system is limited by the maximum value of the material surface load of the settling tank, at which good sludge separation is still ensured in this tank. If there is sufficient capacity of the aeration system in the system, the sludge concentration in the system can be increased by creating a regeneration zone at the beginning of the denitrification zone and redirecting the wastewater supply downstream of this regeneration reactor. In the regeneration volume, the concentration of the sludge will increase to the level of the return concentration, or of excess sludge, thus the age of sludge in the system also will increase. Another possibility for increasing the concentration of sludge in the biological system for increasing the substance load of the settling tank is the application of biofilm biomass carriers in the reactors, which will result in the creation of a hybrid biological system with suspended and attached biomass. This is, for example, an activated sludge process with a built-in biomass carrier (solid plastic structure placed above aeration devices or a partially submerged rotary carrier) or an activated sludge process with a floating biomass carrier (e.g., free-floating polyurethane cubes with 1.0 to 1.5 cm edges or with solid suspended cords). An important requirement for carrier materials is the long-term stability of their properties in an activated sludge environment. Nitrifying and denitrifying volumes usually consist of sections that are connected in series, creating a concentration gradient that ensures the kinetic selection of flock-forming bacteria. In areas with significant temperature changes, it is advisable to ensure the last denitrification and first nitrification sections by mixing and supplying oxygen, which increases the flexibility of the system in terms of their alternative operation in oxic conditions (strengthening the nitrification process in the winter) or in anoxic conditions (increasing the efficiency of the denitrification process in the summer periods when there is sufficient activity of nitrifying bacteria). The majority (60–80%, a value of 75% is usually used in design calculations) of heterotrophic microorganisms in activated sludge can denitrify in anoxic conditions. During the denitrification of 1 mg of NO_3-N, 2.9 mg of O_2 is saved, which is theoretically about 63% of the consumed oxygen, which is consumed for the oxidation of 1 mg of NH_4-N. During the denitrification of 1 mg of NO_3-N, 0.06 mmol of OH^- is released, which prevents a drop in the pH. A necessary requirement for the course of the denitrification process is enough suitable organic carbon. The recommended values for the BOD_5:NO_3-N ratio are in the range of 3 to 4. In the case of an inappropriate composition of municipal wastewater (low BOD_5:NO_3-N ratio), possible solutions to ensure the necessary organic carbon (BOD_5) for denitrification can be, in addition to using an external carbon source, e.g., the reduction of the retention time in primary settling tanks or their decommissioning, acidification of primary sludge, or thermal hydrolysis of sludge. Possibilities for saving the BOD, which is contained in the supplied wastewater, include, e.g., the replacement of screw pumps and aerated sand traps, possibly covering them (unnecessary aeration of wastewater causes a decrease in the BOD_5 content), and the minimization of the transport of dissolved oxygen from the oxic to the denitrification zone of the reactor by internal recirculation. Since for facultative bacteria, oxic conditions are the most favorable from an energy point of view, then anoxic and anaerobic conditions are the least favorable; when dissolved oxygen is introduced into the anoxic zone, the oxidation of BOD_5 by the aerobic respiration of microorganisms occurs primarily. A possible interpretation is that there will be a reduction in the anoxic volume of the denitrification zone. Thus, dissolved oxygen can be considered a serious inhibitor of the denitrification process. In the MLE system (Figure 1), which is equipped with internal recirculation, the required TN output concentration of 8 mg/L can be achieved. Without internal circulation (the original Ludzack–Ettinger (LE) system) a concentration of 12 mg/L TN could be achieved [27]. The operation of a biological system with complete nitrification is, in the case of sewage WW, accompanied by an approximately 50% increase in the energy required for aeration compared to the removal of organic pollution alone [17]. By the appropriate inclusion of the anoxic zone in the technological line, part of the organic pollution is oxidized by anoxic respiration, which will be reflected in the reduction of the operating costs associated with the energy necessary for the supply of oxygen. In addition, denitrification can theoretically reuse 62.5% of the oxygen consumed for nitrification. However, due to incomplete denitrification, the real possibilities are around 50%, which roughly covers the costs of mixing the anoxic zone and nitrate recirculation [17].

The authors of [28] present the feasibility of biological nitrogen removal (BNR) using a modified Ludzack–Ettinger (MLE) system at a reduced hydraulic retention time (HRT) of 5.5 h. Complete nitrification was achieved at 75–80% TN removal at a temperature of 12 °C. There was also a reduction in the net observed yield of biomass by 28% compared to the full-scale plant (0.31 g_{VSS}/g_{CODr}). Compared to the conventional technology using activated sludge (CAS), the sedimentation properties were also improved (the Sludge Volume Index (SVI) value decreased from 202 mL/g to 97 mL/g). The values of the heterotrophic biokinetic parameters of the CAS and MLE systems were very similar and consistent with published values for primary runoff. The values of the specific nitrification rates at a temperature of 20 °C in the MLE with a 0.14 g NH_4-N/(g_{VSSd}) system were 55% higher than in the CAS [28].

At the end of this section, it should be highlighted that nitrifying and denitrifying bacteria are contradictory in terms of the carbon source for the synthesis of new biomass and energy acquisition. Thus, it is necessary to ensure sufficient oxygen conditions for the nitrification process and enough suitable organic carbon for the denitrification process. Their provision must be seen in the wider context of the WWTP technological line as well as in their mutual connections. The main disadvantage of upstream denitrification is the increase in operating costs related to internal recirculation. In addition to operating costs, internal recirculation represents the risk of bringing oxygen into the denitrification volume.

RDN System

Some shortcomings of the system with upstream denitrification are eliminated by the R-D-N system [31,32]. This system is an extension of the upstream denitrification system with a return sludge regeneration zone (R). To suppress the excessive growth of filamentous microorganisms, the entrance part of the denitrification zone can be set aside as an anoxic selector (Figure 2).

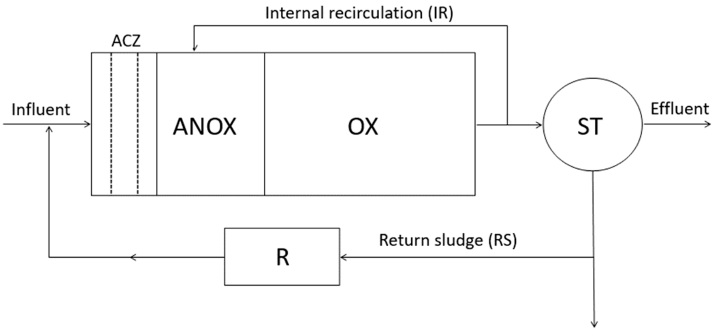

Figure 2. R-D-N system. ACZ—anoxic contact zone, ANOX—anoxic zone, OX—oxic zone, ST—sedimentation tank, and R—regenerator.

This system is characterized by a 20–25% reduction in the required volume compared to pre-denitrification while achieving the same denitrification efficiency. Simultaneously, less internal recirculation is also required. The presence of the regeneration zone ensures the restoration of the accumulation capacity of microorganisms and an increase in the aerobic SRT of the sludge. When controlling aeration in the regeneration zone, it is important to increase the share of simultaneous denitrification at the level of micro-flakes (micro-segregation). Increased phosphorus removal was also observed in this process compared to other nitrification–denitrification systems.

The advantages of the RDN technology are that it not only significantly increases of the age of sludge in the biological system, but also increases the kinetic selection pressure in favor of the growth of well-settling, floc-forming microorganisms. These microorganisms can fill and regenerate the capacity (metabolic selection pressure). The regenerated heterotrophic microorganisms then denitrify faster in the denitrification reactor. Since the microorganisms of the activated sludge are not in contact with the wastewater supplied to the biological stage, nitrification can take place there undisturbed. Sludge water with a high content of ammoniacal nitrogen can be fed into the regeneration reactor, thereby enriching the activated sludge with nitrifying bacteria.

2.1.2. Cyclical Nitrogen Removal

Alternating aerobic and anoxic zones can be achieved in a batch or continuous flow activated sludge system by alternately switching the aerators on and off. This type of intermittent or pulsed aeration in activated sludge facilities is referred to as cyclical nitrogen removal (CNR) [25]. CNR processes can be advantageously applied in existing activated sludge systems for which the permits for nitrogen removal have also been revised. It only requires small process adjustments, e.g., the installation of partitions or timers for the cycling of aeration equipment, pumps to ensure internal recirculation, pipelines, or fittings to ensure a gradual supply. Thus, compared to a single anoxic zone, potential cost savings can be expected when implementing the CNR process.

Simultaneous Denitrification

Nitrification and denitrification processes can take place simultaneously in one bioreactor with spatial or time segregation (macro-segregation level) of oxic and anoxic conditions. Simultaneous denitrification takes place in oxidation trenches and circulation systems with activated sludge in one bioreactor. The creation of oxic and anoxic zones is ensured by the operation of aeration devices. At the same time, the operation of the aerators ensures the sufficient speed of internal recirculation, and thus, the maintenance of the activated sludge in suspension. The process of nitrification and denitrification (SND) can also take place simultaneously in the aerobic reactor. The conditions for the course of these processes are created inside the structure of activated sludge flocs (micro-segregation of oxic/anoxic conditions). The achieved nitrogen removal efficiency without additional carbon is 80 to 96% [5]. The required C:N ratio was 10. The dissolved oxygen (DO) values that need to be ensured are in the range of 0.3 to 0.7 mg/L [34]. The oxidation ditch is an appropriate system for such process. It consists of a ring-shaped channel (Figure 3) about 1–1.5 m deep. A supply of oxygen and the maintenance of activated sludge in suspension are provided by an aerating rotor consisting of a Kessener brush, which is placed across the ditch. The mixed liquor circulates at about 0.1 to 0.6 m/s [35]. Arbitrary flow can be expected due to the sequence of the zones, with different mixing in the bioreactor and an equalization of the impact of pollution and hydraulic loads.

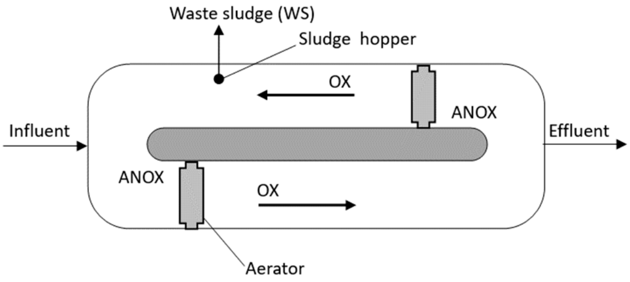

Figure 3. Oxidation ditch.

The oxidation ditch was originally developed for small towns in the Netherlands and is usually operated on the principle of a long-term extended aeration process. The endogenous respiration phase of the growth curve is characteristic for this process. Thus, a relatively low organic load (F/M) and long HRT are maintained. High values of SRT and low excess sludge production are also typical for this mode of operation. Wastewater is usually treated without primary sedimentation. Their main disadvantage is the greater demands on the built-up area. It enables a 90 to 95% BOD_5 removal efficiency. However, in the case of sufficiently long channels (corridors), it enables the creation of oxic and anoxic zones (macro-segregation in space). Controlled aeration can achieve an efficiency for total nitrogen removal in oxidation trenches of around 90% [36].

An oxidation ditch can be operated in the concentration range of 8–12 mg/L of effluent TN [25]. The performance of a ditch system cannot be predicted with as much certainty as other activated sludge systems. An acceptable performance will require field monitoring for optimizing the operation, such as determining the DO profile. The process can then be optimized by adjusting the DO level, which is achieved by turning aerators on or off or by controlling the oxygen transfer rate by varying the aerator submergence and/or horsepower [22].

Intermittently Aerated CSTR

For smaller wastewater treatment plants (e.g., oxidation ditch), it is more appropriate to apply systems with time alternation/segregation of oxic and anoxic conditions. Time segregation is an analogy of spatial segregation. An intermittent operation regarding the oxygen supply can be applied in a batch/semi-continuous mode or a continuous stirred-activated sludge reactor (CSTR). Sedimentation is carried out in a separate sedimentation tank (Figure 4) [36].

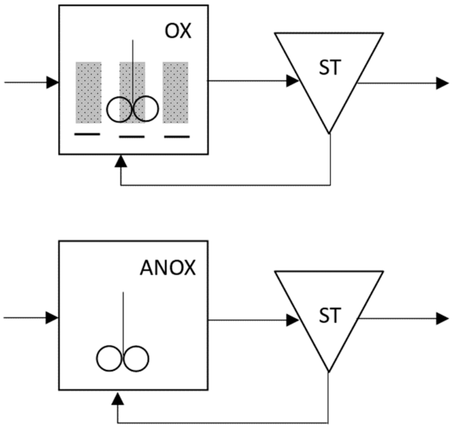

Figure 4. Intermittently aerated CSTR. OX—oxic, ANOX—anoxic, and ST—sedimentation tank.

In continuously operated systems with time segregation, the activation tank is constantly stirred to prevent sludge settling. Based on a suitable signal (like systems with intermittent operation), air is intermittently supplied to the activated sludge system. During aeration, the concentration of nitrates increases, and the concentration of ammonia decreases. In the anoxic period, the course is the opposite [37]. This mode of operation is suitable to use, e.g., in the intensification of small oxidation trenches for the purpose of nitrogen removal [36], where due to the small lengths of the channels, spatial segregation of oxic and anoxic conditions is not possible. Unlike the more often used sequential batch reactor (SBR), described in the next section, this system is close to an ideally mixed system. At low load values, it is necessary to extend the anoxic period to prevent sludge bulking [38]. Intermittent aeration in such a reactor is used intensively for small wastewater treatment plants regarding nitrogen removal [36,38,39].

An important feature of the CSTR systems with time segregation is the high flexibility of changing the length of the cycle or lengths of the oxic and anoxic periods. The common trait of intermittently aerated/operated systems is the possibility of fully automated operation, which creates real conditions for expanding their practical applications.

Sequential Batch Reactors (SBRs)

SBRs are single reactors that cycle through anaerobic, anoxic, and oxic conditions to achieve biological nutrient/nitrogen removal. This technology is particularly suitable where hydraulic and organic loads are highly variable and skilled operating and maintenance personnel are limited [40]. The predecessor of this bioreactor was a batch bioreactor for BOD removal, known as the fill-and-draw process. The reactor was filled with settled wastewater and aerated long enough to oxidize most of the BOD. The contents of the reactor were then allowed to settle, and the treated supernatant was discharged into a receiving water body. Part of the settled sludge was withdrawn, and the whole process was repeated. However, this reactor requires a lot of control, and it is often not used anymore.

Operating an SBR in a semi-continuous mode is inefficient in terms of nutrient removal. Already in the first phase, BOD_5 is removed, which then leads to substrate limitation of the denitrification process. In the case of operating one bioreactor, the risk of increasing the pollution of the treated wastewater by inflowing wastewater in the phases of sludge sedimentation and decantation of the purified wastewater is also a problem. The solution is dosing and repeated feeding of the raw wastewater during the anoxic phase within one operating cycle. With the development of microprocessor control, a modification of this process is gaining more and more popularity. It allows users to carry out several time-segregated processes such as nitrification and denitrification in the same reactor. Since, in addition to individual biochemical reactions, all other operations (filling WW, removing supernatant, thickening sludge, and removing excess sludge) also take place in the same reactor, two or more reactors are needed for the wastewater treatment. Due to the existing concentration gradient and the controlled alternation of anoxic and oxic conditions, a sufficient decrease in the concentration of organic pollution is usually achieved before the oxic period. Thus, the anoxic period fulfils the function of a selector zone [17]. The SBRs can be operated with less than 8 mg/L TN in an effluent when optimizing the DO level [25]. SBRs are convenient for relatively small flows with a high variability.

After dosing the wastewater, denitrification takes place in the anoxic phase. The result of the repeated dosing of wastewater is high values of BOD_5 content, which cause a kinetic selection pressure in favor of strengthening the growth of floc-forming microorganisms. The efficiency of denitrification increases with the amount of dosing of raw wastewater. A system operated in this way combines the advantages of an SBR and upstream denitrification with a high total recirculation ratio (R) (Equation (7)). Increasing the number of doses, therefore, has a similar effect as increasing the total recirculation ratio (R).

Carrousel System

Challenges to the development of the Carrousel process (Figure 5) were the excessive surface area, and therefore, also space requirements, the enormous capital expenditures for surface aerators, and the associated energy costs of designing large-capacity oxidation trenches. The Carrousel system has been developed to provide sufficient aeration and maintain adequate flow rates while maximizing surface area utilization [25]. This type of activated sludge process represents an innovation of an oxidation ditch with respect to a specific kind of aeration. Vertically mounted mechanical aerators are used to introduce oxygen into this system. At the same time, they provide a sufficient horizontal velocity for the activated sludge. Thus, they prevent solids from settling in the reactor channels [41].

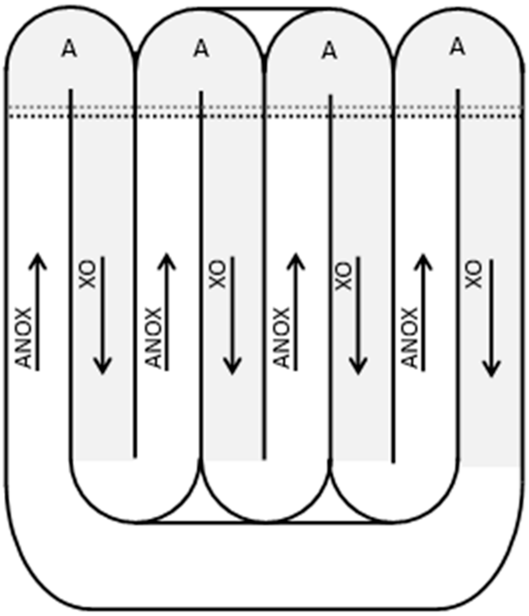

Figure 5. Carrousel reactor. ANOX—anoxic, OX—oxic, and A—aerator.

Technologically it is an extended activated sludge process, for which a high solid retention time (SRT) and hydraulic retention time (HRT), low food-to-microorganism ratio values, and low excess sludge production are representative. The Carrousel system corresponds to a series of completely mixed sections with different dissolved oxygen concentrations.

The application of this bioreactor for domestic wastewater treatment (fed with pre-settled sewage and a sludge load $\geq$ 0.1 kg BOD/(kg-$_{MLSS}$.day)) relates to severe bulking problems (mainly the presence of *Microthrix parvicella*) independent of the type of selector (plug flow or completely mixed) used [42]. This gives further evidence about completely mixed flow in the bioreactor.

The nominal capacity of this bioreactor is significantly higher in comparison to an oxidation ditch. The other advantage of this type of activated sludge process relates to the aerobic stabilization of the activated sludge and the possibility of the exclusion of a primary sedimentation unit in WWTPs. A high control flexibility is typical for this type of bioreactor by changing the number of aerators in use, the rotating velocity of the aerators, and/or the depth of submerging [43].

2.1.3. Multi-Anoxic Zone Systems

The current approach should be towards the use of systems with low oxygen and carbon contents, which are beneficial for saving energy and preventing environmental pollution. The four-stage Bardenpho process (Figure 6) has demonstrated its viability in the reconstruction of existing WWTPs in Chesapeake, for which the permit was revised with nutrient removal requirements [20]. If a sufficient volume of the activation tank is available, its modification to the Bardenpho system may only require the installation of baffles and devices for internal MLSS recirculation. In the case of activation devices in which nitrification was not operated, it will also be necessary to increase the aeration capacity [25,44]. The multi-anoxic zone with a step-feed arrangement (Figure 7) makes it possible to eliminate some of the disadvantages of pre-denitrification. Recirculation with return sludge is usually sufficient. The efficiency (E, %) of denitrification depends on the number of anoxic–oxic sections n (n = 2–4) according to the following relationship (8):

$$E = 1 - \frac{1}{n(RS+1)}100\% \tag{8}$$

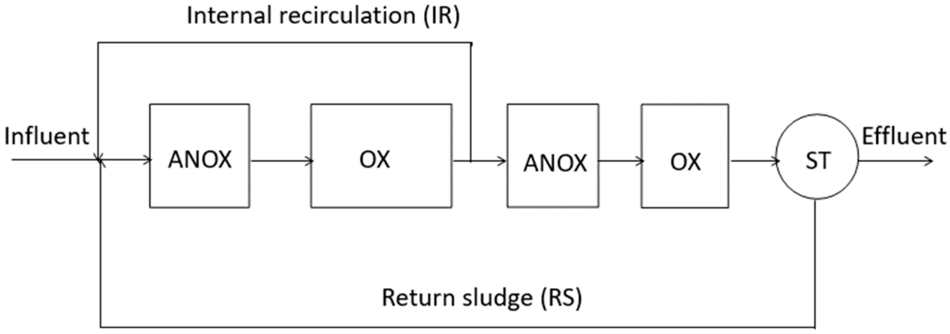

Figure 6. Four-stage Bardenpho system.

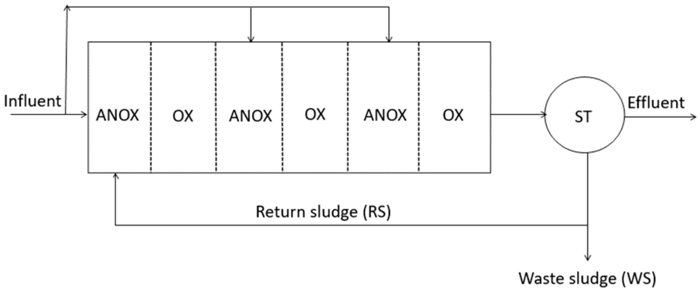

Figure 7. Muti-anoxic zone with step feeding.

With a return sludge recirculation ratio of RS = 1, a 75% denitrification efficiency can be achieved in the four-stage Bardenpho system (Figure 6) in comparison to the multi-anoxic zone with step feeding (Figure 7), where even an 83% efficiency could be achieved. In the case of the multi-stage system (Figure 7), a recirculation ratio of RS = 1 corresponds to a denitrification efficiency of 83.3%. The same denitrification efficiency can be achieved in the MLE system (pre-denitrification, Section 2.1.1) with a total recirculation ratio of R = RS + IR = 5. For higher denitrification requirements, internal recirculation can also be included in six multi-stage systems (Figure 7). These modifications also lead to a gradual equalization of changes in the acid-neutralization capacity (total alkalinity), and thus, also the elimination of the possible negative impact of a decreasing pH on the course of the nitrification process. This is especially advantageous in the case of wastewater with a high nitrogen content or wastewater with a low buffering capacity. Another advantage is the elimination of the influence of internal recirculation on the intensity of axial mixing in individual sections. Due to the cascade arrangement, this system shows a certain concentration gradient. A significant advantage is the smaller total required volume compared to upstream denitrification. In systems with separated sludge (separated-sludge denitrification), concentration limits of <3 mg/L TN can also be achieved. If the strict limit for TSSs is <10 mg/L, separate-stage denitrification using a downflow filter is required. If effluent filtration is not required, an upflow, packed-bed denitrification bioreactor can be used. During the warmer period, it is possible to use cyclic aeration in the nitrification stage to achieve the conditions for denitrification and thereby also reduce the consumption of supplied organic carbon in the denitrification bioreactor, saving on operating costs [25].

With the required output concentrations of 6 to 8 mg/L TN, it is advantageous to use systems with dual anoxic zones. For example, the Bardenpho system makes it possible to achieve output concentrations of <3 mg/L TN in combination with filtration. For the area where effluent concentrations must be in a range of 6–8 mg/L TN, the application of the Bardenpho system with two anoxic zones without effluent filtration [25] is sufficient.

2.1.4. Biofilm-Based Processes

Technologies such as the Submerged Aerobic Fixed Film Reactor (SAFF), trickling filters, and biofilters are commonly known as fixed-bed processes in wastewater treatment. A study of nitrogen removal from a wastewater plant using a submerged attached growth bioreactor showed an 85% average total nitrogen removal [45]. A recent study showed that nitrogen removal of 52–54% was achieved with partial nitritation in sponge trickling filters (STFs) [46]. The report also shows an increased nitrogen removal of more than 60% in currently designed trickling plants.

Andraka et al. [47] used natural zeolite as a carrier of attached biofilm biomass for the intensification of a conventional municipal wastewater treatment plant with activated sludge. The results of the operation of this pilot hybrid system show that the zeolite, with its specific surface, contributed to a significant increase in the content of the biocenosis compared to the system with activated sludge. In the hybrid reactor with a zeolite bed, which represented 20% of the total volume of the bioreactor, significantly higher removal efficiencies were observed for all investigated parameters. The average value of the BOD_5 removal efficiency was 7.8% higher at an output value of 4.8 mg/dm^3. Similar results were also obtained for the total suspended solids (TSSs). At an average ammonia nitrogen output concentration of 3.3 mg/dm^3, a 28.8% higher efficiency of its removal was achieved. The average output value of phosphate phosphorus of 1.2 mg/dm^3 corresponds to an increase in the efficiency of its removal by 26.4%. The measured average values of the output concentration of nitrate nitrogen or of nitrite nitrogen were 3.8 mg/dm^3 or 0.03 mg/dm^3. These results are a good example of the effectiveness of the intensification of the conventional biological stage at the municipal wastewater treatment plant to a hybrid system of suspended and biofilm biomass using zeolite [4] in terms of protecting the receiving water from eutrophication.

2.1.5. Hybrid Systems

In single-reactor hybrid systems with combined suspended and attached growth biomass, a block filling installed in the supporting structure above the aeration elements can be used as a biomass carrier. Another option is the use of submerged modules made of plastic mesh or submerged rotating discs. The growing biomass in the bioreactor can also be fixed in suitably shaped plastic particles that move freely in the MLSS. It is advantageous to use a mesh made of foam plastics, as the biomass is captured not only on the surface of the carrier particles, but also inside the porous structure. This significantly increased the amount of fixed biomass compared to carriers with a solid surface.

In Germany, the Linpor system was developed for the purpose of intensification of overloaded treatment plants [48]. Polyurethane foam cubes are used as a biomass carrier. The main function of the carrier is to concentrate the biomass in the tank without increasing the material load of the settling tank. The amount of carrier can vary from 10 to 40% depending on the required biomass concentration.

As a result of the operational verification of the Linpor system at the WWTP, where the activated sludge system was fed by WW from the brewery and dairy, a more than two-fold increase in the biomass concentration was determined. A significant decrease in the sludge volume index (SVI) values (from 500 mL/g to 85 mL/g) was also measured. These changes occurred 30 days after the addition of polyurethane cubes (20% of the tank volume). Nitrification also started. The nitrogen balance showed that 63% of the oxidized forms of nitrogen (created by nitrification or present in WW) were removed by simultaneous denitrification in the anoxic cores of the carrier cubes.

Another hybrid system is the Captor system, developed by the University of Manchester [49]. The bioreactor is operated on the principle of a pseudo-fluidized layer. The fluidized state of the biomass carrier made of polyester foam is achieved by compressed air distributed by a system of nozzles at the bottom of the reactor. Part of the carrier is regularly removed from the fluidized layer using a mammoth. After straining, the polyester foam particles are mechanically wrung out. The biomass obtained in this way contains up to 6% dry matter. Cleaned particles of the biomass carrier are returned to the reactor using the mammoth. The average concentration of biomass in the reactor is 8 to 12 kg.m^{-3}. For municipal wastewater, the retention time in the system is up to 60 min.

The intensification of the processes of nitrification and denitrification can also be achieved by applying a hybrid system with combined growth and suspended biomass [4]. The use of growth biomass carriers can achieve a two- to three-fold increase in the biomass concentration in the system [48], and subsequently, a proportional reduction in sludge loading. In this context, it should be emphasized that the above-mentioned increase in the biomass concentration will not be manifested by an increase in the loading of the secondary sedimentation tank. The topicality of the application of these systems is also increasing in connection with the need for the increased removal of nutrients. Especially with some industrial wastewaters, a negative to toxic effect of higher concentrations of organic pollution on the nitrification process can be expected. Drtil et al. [50] measured, with 1.5 cm polyurethane cubes, a maximum concentration of 8 mg/L O_2 in the system, at which simultaneous denitrification was still taking place. At the same time, the use of a growth biomass carrier reduces the denitrification volume and thereby also shortens the residence time of nitrifying bacteria outside of oxic conditions. In these systems, an improvement in the sedimentation properties of the sludge compared to the suspended biomass itself is usually observed because of the fixation of filamentous microorganisms in the growing biomass.

It should be highlighted that by using growth biomass carriers, two biocenoses with different sludge ages are cultivated in the system, which enables the simultaneous course of slower biochemical processes (nitrification and removal of slowly decomposable organic substances) as well as faster processes (removal of easily decomposable organic substances). In addition, depending on the carrier used, there is a certain degree of so-called meso-segregation, subsequently creating conditions for simultaneous nitrification and denitrification processes in the growing biomass.

2.1.6. Advanced Biological Nitrogen Removal Systems

The biological processes anammox [51], nitrification and denitrification [52], and simultaneous denitrification [53] are among the most effective for the removal of nitrogenous substances from municipal WW in terms of the achieved efficiency ($\geq$95%), costs, and an environmentally friendly nature.

One of the crucial factors in the biological removal of nitrogen from WW is the presence of a sufficient amount of biodegradable organic carbon. However, an external carbon source must be added to increase the effectiveness of the processes [54–56] if the organic content of wastewater is too low compared to the needed COD:N ratio (analogous to the COD:P ratio for biologically enhanced phosphorus removal). In general, the addition of an external carbon source (e.g., methanol, ethanol, acetic acid, and glucose), which is necessary to increase the kinetics of these processes [55,57], together with the cost of disposal of the excess sludge produced, represent the most significant costs in the operation of a WWTP [58,59]. However, as a source of necessary carbon, e.g., WWs from the product processes of food, confectionary, milk, and dairy products, thanks to the high content of organic carbon, which is biodegradable, can be used [60,61]; cost savings will be achieved and a lower impact on the environment will be assured. A serious disadvantage could be the insecurity of the qualitative and quantitative characteristics of these WWs, which may be related to the diversity of product cycles [57].

Collivignarelli et al. [62] present results of an innovative biological process applied in a sludge line. One of the goals of their work was the possibility of recovering the acceptable carbon residue (supernatant) in denitrification processes, replacing bought external carbon sources. Full-scale tests using a Thermophilic Alternate Membrane Biological Reactor (ThAlMBR) were carried out for 12 months. The ThAlMBR is an advanced biological membrane system used to lysate and oxidize the redundant biological sludge produced by WWTPs through thermophilic bacteria, under controlled conditions of temperature and aeration. The ThAlMBR was applied both on thickened (TBSS) and digested sewage sludge (DBSS), with intermittent aeration conditions. Using the respirometric tests, an excellent biological treatability of the supernatant by the mesophilic biomass was observed and the denitrification kinetics reached with the supernatant (4.0 mg $\text{N-NO}_3^-/(\text{g}_{\text{VSS}}\ \text{h})$) and was set up similar to those of methanol (4.4 mg $\text{N-NO}_3^-/(\text{g}_{\text{VSS}}\ \text{h})$). Thanks to the analogous results attained on TBSS and DBSS, the ThAlMBR proved to be compatible with different sludge line points, maintaining the important minimization of sludge production in both cases.

Kim et al. [63] investigated the use of organic wastes as an alternative to commercial carbon sources. A food waste-recycling wastewater (FWR) was estimated as a convenient alternative to a commercial carbon source for natural denitrification. Process stabilization was linked to the acclimatization and function of bacterial populations due to the change of carbon source. Stable denitrification performance over a period of seven months was achieved in a full-scale WWTP using FWR as an indispensable carbon source. The denitrification performance was stable, with a mean nitrate junking effectiveness of 97.2%. These results demonstrate that FWR can be an effective external carbon source beneficial in terms of reducing costs and environmental impacts.

Nitritation–Denitrification

The process of nitritation–denitrification is also referred to in the literature as a "nitrite shunt", which limits the oxidation of nitrites to nitrites and enables the reduction of the produced nitrite to N_2 by heterotrophic denitrification. Compared to denitrification, the oxygen consumption and organic carbon consumption are 25% and 40%, respectively. Excess sludge production is also lower [64]. The authors of [45] state that by combining the control of the length of the aeration phase and the concentration of dissolved oxygen at a temperature of 15 °C, a nitrogen removal efficiency of 95% is achieved in an SBR with aerobic granular biomass.

ANAMMOX System

A new technology has been developed for the treatment of wastewater with a high nitrogen content, which uses the process of anaerobic ammonium oxidation (ANAMMOX).

This process is performed by anaerobic ammonia-oxidizing bacteria (AOB), which oxidize half of the ammonia to nitrite. ANAMMOX bacteria (*Candidatus Brocadia fulgida*) use nitrogen nitrite as the final electron acceptor to oxidize residual ammonia to N_2 [50]. In 2014, the authors of [46] from Delft University published their work on simultaneous partial nitritation and the ANAMMOX process in a suspended biomass system.

Advanced biological nitrogen removal processes (BNR), e.g., nitritation–denitrification and ANAMMOX, therefore, compared to current BNR processes, show a significant reduction in the need for oxygen and organic substrates. The ANAMMOX process further reduces the oxygen demand by 60% without the need for an organic substrate [64].

2.2. Phosphorous Removal

The combination of an anaerobic environment (Figure 8), i.e., where strictly neither dissolved oxygen nor chemically bound oxygen is present, with aerobic conditions is necessary to remove abundant phosphate from wastewater. Bacteria Acinetobacter spp. and especially the strain Lwoffi were identified as being responsible for accumulating excess phosphates in their cells.

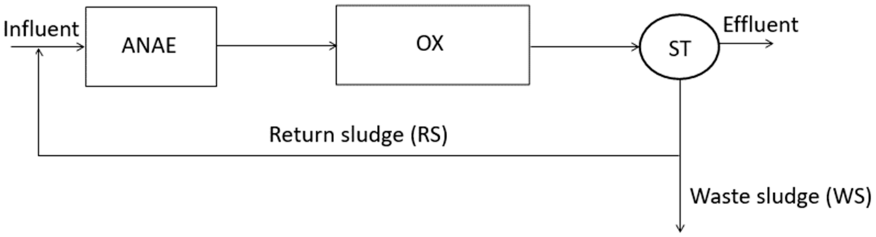

Figure 8. Anaerobic/oxic (AO) system.

Enhanced biological phosphorus removal (EBPR) is performed by microorganisms that can accumulate phosphates (PAOs). This process requires a sequence of anaerobic and anoxic/oxic conditions [65]. PAOs in anaerobic conditions assimilate volatile fatty acids (VFAs) to polyhydroxyalkanoates (PHAs) inside the cells. They obtain the necessary energy by the hydrolysis of intracellular polyphosphate (poly-P). During hydrolysis, orthophosphates are released into the activation mixture. Intracellular glycogen is also transformed into PHAs. Thus, the PHA content inside PAOs increases, and the concentration of glycogen and poly-P decreases. PAOs oxidize PHAs under subsequent aerobic/anoxic conditions, producing energy and organic carbon for the synthesis of new cell mass. The released energy is used by POAs to synthesize poly-P bonds inside the cells [66]. PAOs usually store more poly-P in the aerobic/anoxic phase than was released in the anaerobic phase [67]. The accumulated poly-P is removed from the system by withdrawal of excess sludge under oxic conditions. In recent decades, several technological arrangements have been developed to create a population of microorganisms that is able to remove either nitrogen or phosphorus or both simultaneously [68,69]. The simplest system with an increased biological removal of phosphorus in the mainstream includes a technological arrangement in which an anaerobic zone precedes the aerobic stage. Return sludge is recirculated before the anaerobic stage. This system was named by its originator Barnard as Phoredox. With such an arrangement, bacteria that can accumulate polyphosphates (poly-P bacteria) have the opportunity to synthesize storage substances. In oxic conditions, these are used preferentially for the release of energy, exceeding their needs. Excess energy is used in the formation of polyphosphates and their accumulation in the cells of microorganisms [5,70,71].

The same configuration was patented by Air Products and Chemicals, Inc. under the label ‘Anaerobic/Oxic’, or A/O [17].

2.3. Nitrogen and Phosphorous Removal

2.3.1. Current Methods for Nutrient Removal

The simultaneous removal of nitrogen and phosphorus is the key to improving the quality of secondary effluent of WWTPs to prevent the negative environmental impact in receiving waters, mainly eutrophication. Therefore, it is urgent to develop the applicable technologies for the simultaneous biological removal of nitrogen and phosphorus from secondary effluents. A lot of different techniques were developed recently.

After pilot and full operational trials with the Phoredox system designed for phosphorous removal, Barnard combined this configuration with upstream denitrification [72]. The result of this combination is a three-stage nitrogen and phosphorus removal system (Figure 9). This system was also later patented under the designation 'Anaerobic/Anoxic/Oxic', or A2/O [72].

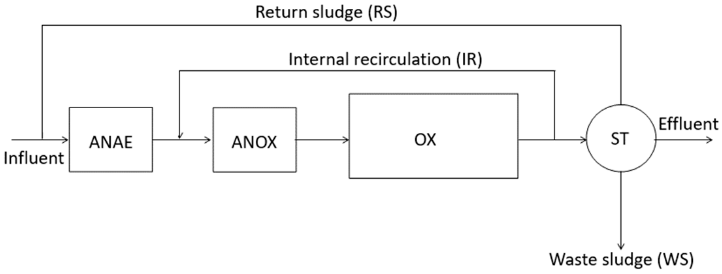

Figure 9. Anaerobic/anoxic/oxic (A2/O) system.

Wanner et al. [73] dealt with the design of efficient process technologies using activated sludge for the efficient removal of N and P from municipal WW. Compared to conventional organic pollution removal technologies, the overall design and optimization of the processes and technologies for nutrient removal are different in many aspects. With sufficiently high values in the temperature, SRT, and sufficient supply of oxygen, it is relatively easy to design and operate the nitrification process. The main problem with nitrogen removal is the lack of organic compounds for denitrification. Even if the amount of usable organic carbon in the supplied WW is sufficient along the technological line of the WWTP, it is lost due to biochemical oxidation (e.g., screw pumps, aerated sand trap, but also the nitrification process and recirculation streams to the anaerobic/anoxic zone). One of the ways of better utilizing the organic carbon and increasing the efficiency of denitrification is inherent to the MLE denitrification system. The recirculation of the stream, which is rich in oxide forms of nitrogen, from the nitrification zone to the denitrification zone, where it is mixed with the incoming WW rich in organic substances, is energy intensive and causes a decrease in the concentration gradients in the technological line. Wanner et al. [73] proposed a technological line (Figure 10) in which organic substances are sequestered under anaerobic conditions and are effectively used as electron donors during the subsequent denitrification. A newly developed modification of the activated sludge system for N and P removal was studied in a laboratory SBR bioreactor to prevent organic substrate losses under oxic conditions. The ammonia contained in the supernatant is oxidized in a biofilm reactor after sedimentation. The nitrate-rich stream is then mixed with activated sludge, which contains sequestered organic substances. In anaerobic conditions, it is possible to separate from WW almost all organic substances and achieve an enhanced biological removal of phosphorus. The ability of poly-P bacteria to synthesize polyphosphates and denitrify under anoxic conditions was experimentally verified.

Another configuration is a five-stage process referred to as "modified Bardenpho" (Figure 11), which was created by combining the Phoredox system (phosphorus removal) and the Bardenpho system (nitrogen removal). The biological stage consists of the following sequence: anaerobic–anoxic–oxic–anoxic–oxic. Return sludge is fed to anaerobic zone. There is one internal recycling stage of nitrates in the system from the outlet of the first oxic zone to the inlet of the first anoxic zone (i.e., from the third to the second tank in the order indicated). In the five-stage Bardenpho system, there should be the possibility of an anaerobic zone in case of more suddenly higher hydraulic loads or in the case of loads with higher concentrations of dissolved oxygen. An alternative option to ensure the operation and control of the process can be a pre-fermentation tank and the division of the anaerobic zone into sections.

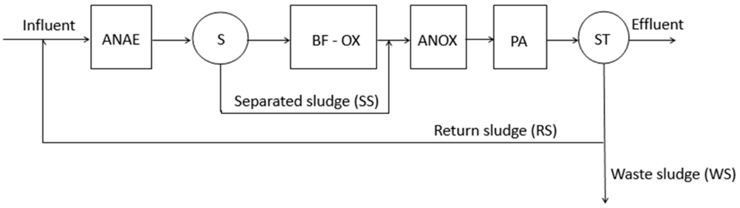

Figure 10. Proposed configuration of the WWTP for nutrient removal.

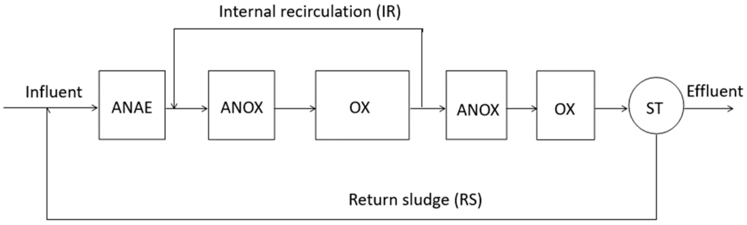

Figure 11. Five-stage Bardenpho system.

Effluent concentrations less than 8 mg/L cannot be consistently obtained using single anoxic-zone processes without an additional attached growth filter, or effluent concentrations of the external organic carbon supplement TN less than 6 mg/L can be practically attained in a suspended growth system without external organic carbon addition by placing an endogenous anoxic zone in series after the aerobic zone. While the A2/O process does use two unaerated zones, the first (anaerobic) zone is not used for enhanced nitrogen removal but is provided for phosphorous removal or as an anaerobic selector. The first documented case of a second anoxic zone for denitrification was credited to Barnard, depicted schematically in Figure 9. This process served as a precursor to the process he later patented as the Bardenpho process. Phosphorous removal was later accommodated in the Bardenpho process by placing an anaerobic reactor at the head of the treatment train, resulting in a five-stage process, which is also illustrated in Figure 11. The anaerobic fifth stage can be included in facilities that are not required to remove P as an anaerobic selector to suppress the growth of filamentous organisms. During the 1980s, other alternatives for the simultaneous biological removal of nitrogen and phosphorus were developed, mainly initiated by the knowledge of the negative impact of nitrates on the biological removal of phosphorus due to the use of an easily decomposable substrate by nitrifying bacteria. On this basis, the three-stage Phoredox process was modified at the University of Cape Town to eliminate nitrate recycling to the anaerobic tank in the return sludge. This process is known as University of Cape Town (UCT) and is schematically shown in Figure 12.

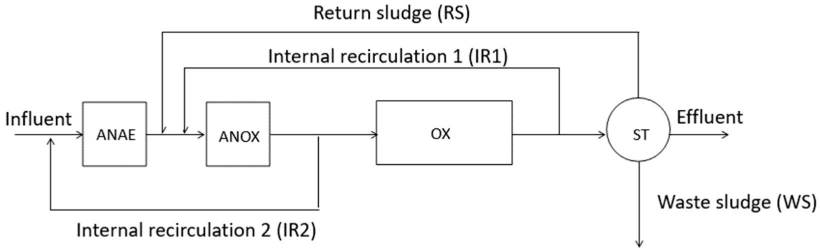

Figure 12. UCT process.

The UCT process was also further modified by providing two anoxic zones (instead of one as in the original UCT) and two separate internal recycle lines. The purpose of this modification was to control the return sludge (RS, shown in Figure 13) and nitrate recycling separately and to reduce the NO_3 load to the anaerobic reactor. Although the Modified UCT process uses dual anoxic zones, the second anoxic zone is not an endogenous denitrification reactor as is described for the Bardenpho process. Instead, the second anoxic zone in the Modified UCT is used only to denitrify recycled nitrates from the aerobic zone, and the first anoxic zone is exclusively used as an exogeneous denitrification reactor to denitrify the RS before recycling to the anaerobic zone. This allows for increased recycling rates to the second anoxic zone for denitrification and reduces nitrate interference of P removal in the anaerobic reactor.

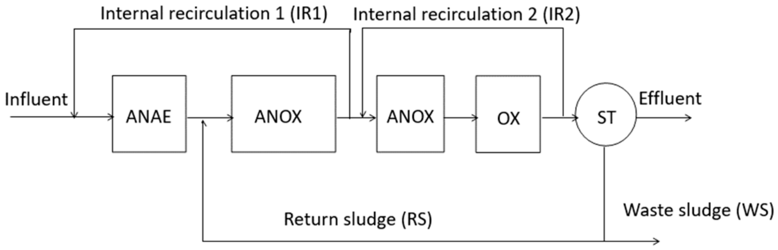

Figure 13. Modified UCT process.

The Virginia Plant Initiative (VIP) process was developed and patented by Hampton Roads Sanitation District and CH2M Hill [74]. In the VIP method (Figure 14), bioreactors are the same as in the A2/O system. However, the returned activated sludge is recycled to the anoxic zone, and the MLSS is recycled from the anoxic zone to the anaerobic tank (IR2). Nitrate is recycled from the oxic zone to the anoxic tank for denitrification (IR1), thus reducing the amount of nitrate fed to the anaerobic zone in return-activated sludge (RS). Heidari [75] used BioWin software (6.2.10.3739) to design, upgrade, and optimize wastewater treatment plants by comparing nutrient removal efficiencies between the A2O and VIP processes. The results of simulations showed that even though both the VIP and A2/O achieved excellent nutrient removal, the VIP could remove more nutrients than A2/O.

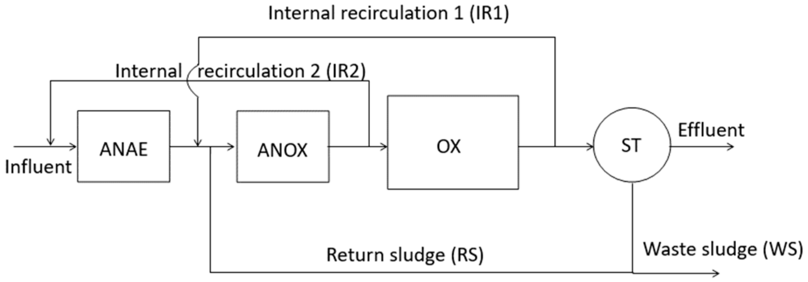

Figure 14. VIP process.

Wanner et al. [73] present a brief overview and basic characteristics of activated sludge systems for removing N and P from wastewater. On a pilot-operational scale, they studied, using real wastewater, four modifications of the process using activated sludge for N and P removal. The results showed that the aeration of the returned sludge significantly contributed to achieving a high efficiency of N removal. In the design of the new municipal wastewater treatment plant, the process of return sludge aeration was used, which consists of anaerobic, denitrification, and nitrification zone sequence. This newly designed R-AN-D-N technology (Figure 15) is characterized by great flexibility, which is related to the supply of WW to both anaerobic and anoxic zones, as well as the possibility of introducing internal recycling in the denitrification or anaerobic zone. In terms of development, it is an extension of the RDN system with a regenerator (R). Four modifications of the activated sludge process (pre-denitrification (DN), RDN process, modified Phoredox process, and R-AN-D-N process) for nutrient removal were tested. The best nitrogen removal was observed in the RDN system. The most stable and best results of phosphorus removal were achieved in Phoredox-type processes. All systems had good sedimentation and thickening properties. A significant improvement in the sedimentation and thickening properties of sludge was characteristic in systems with an incorporated anaerobic zone. The results of testing the alternative R-AN-D-N were used for the design of a real municipal wastewater treatment plant.

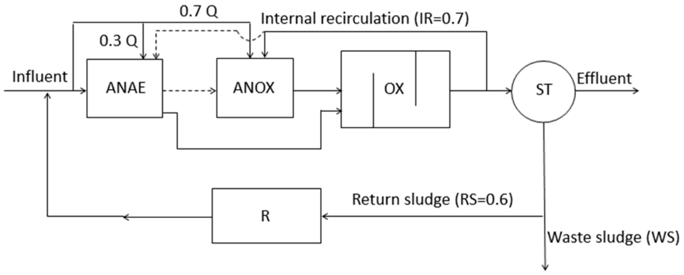

Figure 15. The technological line of the R-AN-D-N process.

The processes and microorganisms occurring in biological nutrient removal are summarized in Table 1.

Table 1. Processes and microorganisms occurring in biological nutrient removal.

Nutrient and Form		**Process**	**Process Description**	**Bacteria**	**References**
Nitrogen	Ammonium	Nitritation (1st step of nitrification)	Ammonium nitrogen is transferred to **nitrite** in aerobic environment by chemoautotrophic nitrification bacteria.	*Nitrosomonas*	[17]
		Annamox	Ammonium nitrogen is transferred to **nitrate** and **nitrogen gas** by chemolithotrophic microorganisms in anaerobic environment.	*Candidatus Brocadia*	[21,22]
	Nitrite	Nitratation (2nd step of nitrification)	In an aerobic environment by chemoautotrophic nitrification bacteria, nitrite transferred to **nitrate**.	*Nitrobacter*	[17]
	Nitrate	Denitrification	Nitrate nitrogen is utilized as the terminal electron acceptor, releasing **nitrogen gas** as the product in the process of denitrification.	*Pseudomonas*	[17]
	Ammonium, nitrite, and nitrate	HNAD (heterotrophic nitrification and aerobic denitrification)	Many of these heterotrophic nitrifiers are able to denitrify their nitrification products, i.e., nitrates and/or nitrites, to nitrogen gas.	*Marinobacter*	[76]
Phosphorous	Phosphate	EBPR (enhanced biological phosphorus removal)	The process requires a sequence of anaerobic and anoxic/oxic conditions and is provided by microorganisms that are able to accumulate phosphates.	*Acinetobacter*	[64]
Phosphorous and Nitrogen	Phosphate, nitrite, and nitrate	SNDPR (simultaneous nitrification–denitrification–phosphorus removal)	Remove P with oxygen, nitrate, or nitrite in oxic and anoxic environments.	*Accumulibacter, Thauera*	[74,77]
Phosphorous Nitrogen	Phosphate, nitrite, and ammonium	HNAD (heterotrophic nitrification and aerobic denitrification)	Remove nitrogen through heterotrophic ammonium nitrification coupled with fast nitrite denitrification, phosphorous in anaerobic–anoxic conditions	*Pseudomonas stutzeri YG-24*	[67]

Usually, an effluent concentration of 8 mg/L TN is presented as the performance of single anoxic-zone systems such as A2/O, UCT, and VIP systems. However, concentrations in the range 6 to 8 mg/L TN can be maintained with flexible operational abilities. The decrease in the TN value limit must be compensated with an enhancement of the internal mixed liquor recirculation from the oxic to anoxic zone. Internal recirculation ratio from 1 to 4 should be provided [25]. An additional denitrification zone using an external carbon source can be used to remove low nitrate concentrations. With the required output concentrations of 6 to 8 mg/L TN, it is advantageous to use systems with dual anoxic zones. For example, the Bardenpho system makes it possible to achieve output concentrations of <3 mg/L TN in combination with filtration. For an area of output concentrations of 6–8 mg/L TN, the application of the Bardenpho system with two anoxic zones without effluent filtration [25] is sufficient. In systems with separated sludge (separated-sludge denitrification), concentration limits of <3 mg/L TN can be achieved. If the strict limit for TSSs is <10 mg/L, separate-stage denitrification using a downflow filter is required. If effluent filtration is not required, an upflow, packed-bed denitrification bioreactor can be used. During the warmer period, it is possible to use cyclic aeration of the nitrification stage to achieve the conditions for denitrification and thereby also reduce the consumption of supplied organic carbon in the denitrification bioreactor, saving on operating costs [25]. If moderate levels of TN removal are required, single sludge, single anoxic-zone systems such as the A2/O, VIP, and UCT processes can be selected for phosphorous removal. These systems can maintain effluent WWs with less than 1 mg/L TP. However, TP concentrations in the effluent are variable. The choice between the A2/O, VIP, and UCT processes is mainly influenced by the characteristics of the WW. A reliable performance of less the or equal to 1 mg/L TP will require chemical precipitation in addition to biological P removal. As discussed previously, dual anoxic-zone systems are typically applied where low levels of TN are also required. In this case, the modification or five-stage Bardenpho system would be considered, since it includes an anaerobic zone at the beginning of the process line. Effluent TP levels of less than 3 mg/L have been reported with these systems [25]. To obtain a TP limit less than 2 mg/L and consistently maintain an effluent with less than 2 mg/L TP, chemical precipitation should be included in the treatment line. Biological nutrient removal systems for both N and P removal present some unique operating circumstances, since the two processes have conflicting requirements. The mechanisms for biological phosphorous removal in a mainstream technology such as UCT, A2/O, VIP, or a modified Bardenpho system is through the assimilation of enhanced levels of P in the bacteria selected by anaerobic zone and subsequent elimination of P through waste sludge. Thus, biological P removal is optimized at a shorter solid retention time (SRT) than is typically applied for nitrification. Sludge production and subsequent P removal is promoted at shorter SRTs. Generally, the compromise is to operate the systems at the shortest SRT required to achieve effluent nitrogen limits [25]. Nitrates also present a problem for biological phosphorous removal processes. For enhanced P removal to proceed, the bacteria must first assimilate organic substrate in the anaerobic zone and store them within their cells. Nitrates that may be present in the RAS allow facultative heterotrophic bacteria to compete for the soluble substrate required by the P-removing bacteria. This can degrade the phosphorous removal performance of systems as the A2/O, or modified Bardenpho system with recycled RAS directly to the anaerobic zone. The UCT and VIP processes avoid this problem by returning the RAS to the anoxic zone, where the nitrates are reduced by denitrification. Mixed liquor is than recycled from downstream of the anoxic zone to the anaerobic zone [25]. A key criterion for BNR systems to remove both N and P is the $TBOD_5$:TP ratio. If this ratio is larger than 20:1, than the recycling of nitrates to the anaerobic zone may not be a problem, because there is an adequate supply of excess organic substrate. In this case, either the A2/O or modified Bardenpho system should be considered for stricter limits on the TN. These systems do not have the costs associated with additional recycling from the anoxic to the anaerobic zone. If the $TBOD_5$:TP ratio is less than 20:1, then the VIP or UCT process should be considered. For the same reasons, the TKN:BOD is important.

A high TKN:BOD may indicate that there is insufficient carbon to denitrify completely, resulting in a carryover of nitrates to the anaerobic zone [25]. For WW treatment plants that must remove P and are required to nitrify but not denitrify, it may still be advantageous to include denitrification as a component of single-sludge system. This will reduce the interference of nitrates with the biological phosphorous removal mechanisms and will also provide the cost savings associated with the return of alkalinity and reduction in aeration requirements. A mainstream biological phosphorous-removal system will require effluent filtration to achieve lower levels of TP than 0.5 mg/L. This is because the solids that are carried by the clarified effluent will contain an enhanced level of phosphorous, which can have a significant impact on the effluent TP content. Typically, activated sludge solids will contain approximately 2% of P, whereas the solids from a biological P-removal system will contain typically 4–8% of phosphorous [25]. In the UCT, A2/O, and VIP systems, the concentrations of TN in the range 8–12 mg/L can be met [25]. The UCT or VIP technologies would be convenient in cases when high phosphorous removal is required and the BOD_5:P ratio < 20:1. Both these systems have high rates with a low SRT and short HRT [25]. Usually, the effluent concentration of 8 mg/L TN is presented for the ideal performance of single anoxic-zone systems such as A2/O, UCT, and VIP systems. However, concentrations in the range of 6 to 8 mg/L TN can be maintained with flexible operational abilities. The decrease in the TN value limit must be compensated for with an enhancement in the internal MLSS recirculation from the oxic to anoxic zone. An internal recirculation ratio from 1 to 4 should be provided [25]. An additional denitrification zone using an external carbon source can be used to remove low nitrate concentrations.

The P content in the dry matter of ordinary activated sludge is about 1 to 2%. Sludge that is exposed to alternating anaerobic and oxic conditions can accumulate 5 to 7% P. By removing such excess sludge, the concentration of P in the purified WW can be reduced below 2 mg/L. For the implementation of the increased biological removal of phosphorus, it is necessary to achieve a retention time for the activation mixture of 0.5 to 3 h. This zone needs to be divided into three to four sections. The concentration gradient designed in this way, in combination with anaerobic conditions, creates a kinetic and metabolic selection pressure to suppress the growth of some filamentous microorganisms. Wastewater must contain enough suitable organic substrate. The recommended BOD_5:P ratio is 20. In the case of a low BOD_5:P ratio, it is necessary to minimize BOD_5 losses in the mechanical stage of WW treatment, similarly to the case of upstream denitrification. The value of this ratio can also be increased by fermentation or hydrolysis of the primary sludge before its introduction into the anaerobic stabilization reactor. It is necessary to minimize the recirculation of NO_3-N and dissolved oxygen into the anaerobic reactor. The excess sludge is drawn away from the oxic reactor/zone to prevent it from remaining in anaerobic conditions outside this reactor before it enters the anaerobic stabilization reactor, or before the release of PO_4-P into the sludge water. For the quick thickening of excess sludge, rotary thickeners, centrifugation, or flotation can be used. At low required concentrations of phosphorus (below 1.5 mg/L) in the outflow of the system with an increased biological removal of phosphorus, settling tanks also play an important role. It should be borne in mind that under these circumstances, there is up to 0.5 mg/L of P in 10 mg/L of suspended solids. A combination of increased biological removal and the chemical precipitation of phosphorus is a common option [29].

2.3.2. Advanced Methods for Nutrient Removal

The authors of [5] analyzed information on current and advanced technologies used to remove N and P from domestic WW. They discuss the mechanisms, advantages, disadvantages, and limitations of these technologies. They recommend an integrated approach to nitrogen and phosphorus removal. They consider the process of simultaneous nitrification and denitrification (SND) to be an advantageous method for removing nutrients from domestic WW due to its operational simplicity, cost effectiveness, low energy demand, efficiency, and minimum demand for pH regulation. Phosphorus denitrification removal appears to be a promising process that provides remarkable energy savings, lower sludge production, an increased efficiency, and reduced carbon-source conditions. It can thus effectively use the limited carbon resources present in domestic WW. The integration of DPAO into the SND process is recommended to realize DPAO-mediated denitrification in a simultaneous nitrification–denitrification–phosphorus removal (SNDPR) system. It is also recommended that a combination of archaeal ammonium and anammox-modified denitrification phosphorus removal processes is applied to consolidate nitrogen removal and phosphorus recovery from low carbon/nitrogen wastewaters under normal operation without adding an external carbon source.

Denitrification Removal of Phosphates

Phosphorus-accumulating organisms (PAOs) that mediate enhanced biological phosphorus removal (EBPR) require alternating anaerobic and aerobic/anoxic conditions for optimal phosphate uptake. The effectiveness of such approaches requires separate unit operations and controlled experimental conditions, increasing the operational difficulties and technology costs. However, in the literature, one can find many claims about the existence of some heterotrophic denitrifiers that can remove phosphorus under anoxic/aerobic conditions, i.e., without the need to alternate anaerobic and aerobic/anoxic conditions [77–81]. Heterotrophic microbes are characterized as denitrifying phosphorus-accumulating organisms (DPAOs). These are facultative anaerobic bacteria that can use nitrate and/or nitrite as the final electron acceptor instead of oxygen. The process of phosphorus removal under anoxic/aerobic conditions is also known as denitrification phosphorus removal. Phosphorus removal and denitrification occur simultaneously [79]. According to the literature, both PAOs and DPAOs have enzymes for aerobic metabolism. However, only DPAOs are capable of denitrifying phosphorus uptake. On the other hand, PAOs lack the essential enzymes to utilize nitrate as the final electron acceptor [82]. DPAOs can synthesize polyphosphate by the action of the enzyme polyphosphate kinase (PPK) in the presence of external carbon sources under aerobic or anoxic conditions, i.e., without alternating anaerobic and aerobic/anoxic conditions [83]. Applications of the denitrifying absorbed phosphorous system include some improvements over EBPR methods. These are energy savings, less sludge production [84], and less requirements for the carbon source. DPAOs use the same carbon source for both denitrification and phosphorus removal, resulting in approximately 50% savings in the COD [85]. The simultaneous removal of nitrogen and phosphorus can effectively utilize the limited carbon resources present in domestic wastewater with a moderately low carbon-to-nitrogen ratio [80]. Recently developed advanced biological nutrient-removal (ABNR) technologies create conditions for stable and high-performance N and P removal. They differ from current BNR technologies in several ways [85,86]. They are aimed exclusively at improving the efficiency of BNR, saving energy and operating costs. These technologies include, e.g., simultaneous nitrification/denitrification (SND), bioaugmentation batch-enhanced treatment (BABE), anaerobic ammonium oxidation (anammox), denitrifying phosphorus removal, single reactor for high activity ammonia removal over nitrite (SHARON), etc. [87–90].

Heterotrophic Nitrification and Aerobic Denitrification

Heterotrophic nitrification and aerobic denitrification (HNAD) solve the shortcomings of current biological nitrogen removal systems. It is known that these systems are implemented discretely in two separate anoxic and oxic bioreactors due to the different requirements of the specifics of the individual processes and the microorganisms used (slower speed of autotrophic bacteria, their sensitivity to high organic load, and sensitivity of denitrifying bacteria to the presence of oxygen). Such a solution requires strict compliance with the necessary conditions, which translates into high costs [12]. Recent research studies have demonstrated the existence of some heterotrophic nitrifiers that are able to perform nitrification using organic carbon [91–93]. Many of these heterotrophic nitrifiers are able to denitrify their nitrification products, i.e., nitrates and/or nitrites, to nitrogen gas [94].

These microorganisms can thus use dissolved oxygen and nitrates at the same time and enable nitrification and denitrification processes to be carried out in one reactor under oxic conditions. They use nitrates/nitrites as the final electron acceptor. This process is referred to as the SND process [95]. SND through HNAD provides many advantages compared to current technologies by removing N. It is cost effective, as evidenced by a 22–40% reduction in carbon source consumption and about a 30% reduction in sludge production [77]. The implementation of nitrogen removal processes in one reactor is much simpler. A higher efficiency due to higher growth rates of heterotrophic microorganisms, their ability to use organic carbon as an energy source, as well as the ability to use the products of the nitrification process as reactants for denitrification are reflected in the higher efficiency of this bioreactor. The low DO requirement due to the reduced intensity of aeration and the minimization of recirculation translate into lower minimal energy consumption needs for pH control. The alkalinity generated during denitrification partially balances the acidity generated during nitrification [76]. The variety of substrates and products of heterotrophic nitrification allows for the expansion of application possibilities [96]. The HNAD technology also represents an alternative for the treatment of domestic WW with a low C:N ratio and/or CO_2 limitation for autotrophic nitrifying bacteria [97]. This technology overcomes the limitations of current nitrogen removal technologies and provides a viable method for biological nitrogen removal.

Integrative Approaches for Simultaneous Nutrient Removal and Recovery

It is well known from the literature that SND can offer a one-step treatment process design for nitrogen removal [67,98,99]. As previously mentioned, denitrification phosphorus removal can simultaneously remove nitrogen and phosphorus under either aerobic or anoxic conditions in a single reactor without the alternation of aerobic/anaerobic (anoxic) conditions [100]. Thus, under these conditions, DPAOs can be incorporated into the SND process to develop a simultaneous nitrification–denitrification–phosphorus removal (SNDPR) system. In this process, there is a difference from the conventional enrichment of biomass with phosphorus by the EBPR mechanism to the enrichment of the liquid phase with phosphorus mediated by DPAOs. Phosphorus from the enriched liquid phase can be easily recovered by induced crystallization, which is significantly simpler than as was the case for phosphorus removal and recovery in the EBPR system [101]. Connecting DPAOs to systems for simultaneous nutrient removal and recovery will certainly be the most environmentally friendly and efficient approach. The application of a promising energy- and resource-efficient alternatives to anammox in the mainstream would certainly be successful. However, the selective enrichment of AOB and the simultaneous inhibition of NOB due to a stable supply of nitrites remain a fundamental barrier that prevents its practical implementation [90]. Ammonia-oxidizing archaea (AOA), with their high efficiency of ammonia oxidation under oxygen-limited conditions, are potentially a more suitable microorganism for supplying nitrite and could be an option to link with mainstream anammox [102]. The review presented in [81] proposes another integrated energy-efficient approach by combining archaeal oxidation of ammonium (anammox) with modified denitrification. Due to the reduced carbon-source requirement and the fact that DPAOs use the same carbon source for both denitrification and phosphorus removal, this saves approximately 50% in COD requirements [85]. The simultaneous removal of N and P can effectively use limited carbon sources in domestic WW with a moderately low carbon-to-nitrogen ratio [86].

The potential of advanced biological nutrient removal (ABNR) methods offers a number of advantages over currently used techniques. Despite the abundance of organic matter in wastewater, the effective utilization, including the loss of organic matter, recirculation, excessive sludge formation, and oxygen waste is a persistent problem. However, advanced methods such as simultaneous nitrification and denitrification (SND) together with phosphorus denitrification provide solutions to these problems. ABNR methods maximize the use of limited carbon resources in wastewater, resulting in energy savings, reduced sludge production, and an increased efficiency. The integration of denitrifying phosphorus-accumulating organisms (DPAOs) into SND systems enables the simultaneous removal of nitrogen and phosphorus, improving nutrient recovery and ecological sustainability. In addition, ABNR optimizes resource use by using carbon resources efficiently and minimizing waste. This leads to energy savings, reduced operating costs, and economic sustainability. In addition, these methods promote environmental sustainability by reducing the need for chemical additives and minimizing energy consumption. Another integrated approach is the combination of archaeal ammonium (anammox) oxidation with modified denitrification, where approximately half of COD requirements are saved due to reduced carbon-source requirements. Ease of use is another advantage of ABNR methods that make wastewater treatment processes more efficient and reduce their complexity. These approaches are scalable and adaptable to different treatment scenarios, providing versatile and effective nutrient removal solutions for wastewater.

3. Nutrient Recovery

Nutrients are essential elements for sustaining life, including nitrogen and phosphorus compounds that are crucial for various biological processes [103]. Nitrogen and phosphorus play a key role in the structure and function of living organisms. Nitrogen is an essential component of cellular DNA, amino acids, and chlorophyll, while phosphorus serves as the cells' "energy currency" in the form of adenosine triphosphate (ATP) [104,105]. In agricultural systems, nitrogen and phosphorus play a critical role in promoting plant growth and ensuring the food supply [104].

Despite the abundance of nitrogen in the atmosphere, its availability in the soil is limited, requiring fixation into reactive forms such as amino acids, nitrates, and ammonia to increase its availability for plant uptake [104]. However, natural fixation processes are not sufficient to meet the demands of the growing global population, leading to an increasing trend of anthropogenic production of reactive nitrogen [106]. Conversely, phosphorus faces depletion problems as a non-renewable resource that is primarily obtained from igneous and sedimentary deposits through mining [104]. Current extraction rates indicate that easily exploitable phosphorus resources will be depleted within the next few decades [107]. The importance of nitrogen and phosphorus in agricultural productivity cannot be overestimated. Synthetic fertilizers, composed mostly of nitrogen and phosphorus compounds, have played a key role in bridging the gap between malnutrition and adequate nutrition for a significant portion of humanity [104,108]. However, the inefficient use of nutrients leads to significant losses, with only a fraction of the reactive nitrogen used accumulating in crops, while the rest escapes to the environment through air and water. These losses not only affect human health and biodiversity, but also contribute to environmental problems such as global warming and the eutrophication of water bodies [109].

To address these challenges, increasing emphasis is being placed on nutrient recovery as a means of reducing the load on reactive nitrogen and phosphorus production, and at the same time, mitigating environmental degradation. Nutrient recovery works on the principles of reduce, reuse, and recycle to reintroduce recovered nitrogen and phosphorus as fertilizers to increase agricultural productivity while minimizing adverse environmental impacts. By effectively managing nutrient cycles and reducing nutrient losses, nutrient recovery holds promise for ensuring sustainable resource management, reducing food production costs, and promoting environmental protection [104].

3.1. Ion Exchange/Adsorption

Reactive nitrogen, primarily existing as NH_4^+, is effectively recovered using ion exchange and adsorption-based processes. Zeolite, a popular ion exchanger and adsorbent for nitrogen recovery, offers a tetrahedral framework where aluminum and silicon atoms form interconnected cages and channels [104,110]. The substitution of silicon with aluminum creates a net-negative charge within the zeolite framework, balanced by exchangeable cations such as Na^+, K^+, Ca^{2+}, and Mg^{2+} on the zeolite surface. The performance of zeolites in nitrogen recovery is influenced by various factors such as the pH, influent NH_4^+ concentration, temperature, and ionic strength. Modification techniques, including treatment with acids, alkalis, and salts of alkaline metals, enhance the adsorption capacity and purity of zeolites.

According to a study by Muscarella et al. [111], the zeolite exhibits an adsorption capacity of 19 g NH_4^+/kg, with removal efficiencies ranging from 80% to 95% at pH 6.5. Alternatively, the use of a synthetic resin resulted in the removal of 27.6 g NH_4^+-N/l resin, with a high cation-exchange capacity of 99% achieved after regeneration with HCl [111]. Numerous materials have been proposed as adsorbents for phosphorus. However, each material faces limitations specific to its composition, complicating the determination of universal removal and regeneration efficiencies. Commonly used adsorbents include ferric and aluminum hydroxides, known for their ability to form ligands with oxyanions like phosphates. Recent studies have explored phosphorus sorption onto various substrates such as fly ash, blast furnace slags, gas concrete, crushed oyster shell powder, alum sludge, active red mud, synthetic boehmites, and calcite. Investigating opportunities for nitrogen recovery and reuse becomes essential after zeolites are exhausted. Regeneration techniques, such as NaCl solution regeneration, acid regeneration, regeneration with heat, or biological regeneration, provide concentrated streams of NH_4Cl and $NaNO_3$ in chemical and biological regeneration. In NaCl solution, the NH_4^+ is desorbed and exchanged with Na^+ in solution. In these concentrated regenerant solutions, after raising their pH above 9.3, NH_3 can be air-stripped off, resulting in NH_3 gas that can be further processed for fertilizer production [104]. Ion exchange processes are also effective for removing and recovering phosphorus from wastewater, typically through adsorption onto phosphorus-selective media. These media can exchange anions like Cl^- with PO_4^{3-} ions, producing phosphorus-depleted effluent while maintaining charge neutrality in the solid media [112]. The advantages of ion exchange include high selectivity for NH_4^+, high removal, fast uptake kinetics, simple application and operation, and its ability to work in varying wastewater compositions, flow rates, and temperatures. However, drawbacks include poor selectivity towards PO_4^{3-}-P, inefficient regeneration, and loss of loading capacity [110,113].

To maintain the efficiency of ion exchange media, periodic regeneration is necessary, adding to the operational costs of water treatment. Ion exchange treatment is suitable for removing phosphorus from effluents with concentrations below 2000 mg/L. Despite the promising potential of using zeolites for nutrient recovery, the process remains costly, particularly for large-scale wastewater treatment plants [110].

3.2. Ammonia Stripping

Ammonia stripping, a pH-dependent process, is widely used for nitrogen removal in wastewater treatment. At a pH of around 9.3, the ammonium nitrogen in the solution is converted to ammonia gas, which is facilitated by the addition of lime or caustic soda to maintain the pH around 10.8–11.5. The process involves the conversion of ammonium ions to ammonia in solution. At the same time, the introduction of air facilitates the conversion of ammonia into ammonia gas and removes it, as shown in the reaction (9) below [104]:

$$NH_4^+ + OH^- \overset{air}{\rightarrow} H_2O + NH_{3(g)} \tag{9}$$

This conversion allows for the efficient removal of ammonia gas by air stripping, especially in packed towers that provide a large surface area for mass transfer. The ammonia stripping efficiency is affected by a variety of factors, including the ammonia concentration, hydraulic loading, air flow, pH, and temperature. Studies have shown that increasing the pH increases the efficiency of ammonia recovery up to 92%, with the optimum pH range typically between 8 and 11. Another study report that ammonia removal efficiencies range from 80% to 96.6% at optimal pH and aeration times, highlighting the effectiveness of the process in recovering nutrients from wastewater streams. It was also concluded that air stripping works independently of pH levels when temperatures exceed 80 °C. Similarly, the air stripping time also affects ammonia removal, with optimum removal being achieved within specific time frames [104]. Optimization studies by Guo et al. [114] showed a significant increase in the ammonia removal efficiency with pH values up to 11. Furthermore, the increase in removal was not substantial. Air stripping time optimization studies showed that an 18 h duration resulted in an optimal ammonia removal efficiency [114]. Recent advances in ammonia removal include the development of innovative gas–liquid contactors, such as water jet aerocyclones (WSA), which exhibit a higher stripping efficiency and superior mass transfer efficiency compared to traditional methods such as stripping tanks and packed towers. These improvements help reduce the operating costs and improve the overall processing efficiency.

Ammonia stripping has been used successfully in large-scale treatments in municipal wastewater treatment plants, offering an effective solution for nitrogen removal. However, issues such as the pollution of loaded towers and scaling issues need to be addressed to ensure continuous and efficient operation [115].

3.3. Membrane Separation

Membrane separation processes have emerged as key technologies in wastewater operations, offering the efficient and selective removal of nitrogen and phosphorus compounds from various wastewater and liquid waste streams. Membrane nitrogen recovery processes offer significant advantages, including independence from gas or liquid flow rates, the absence of secondary pollutants in the ammonia permeate, and consistent efficiency unaffected by changes in the ammonia concentration [104]. Nunes and Peimmann [116] demonstrated the regeneration of gaseous ammonia in an acidic solution using a gas-permeable membrane operated under vacuum pressure. Hasanoglu et al. [117] achieved the recovery of ammonia as $(NH_4)_2SO_4$ on the permeate side of a macroporous hydrophobic membrane. Kurama et al. [118] reported a substantial 96.9% recovery of ammonium ions using reverse osmosis (RO). Gerardo et al. [119] used crossflow microfiltration (MF) to recover nitrogen and phosphorus from dairy farm sludge. Mondor et al. [120] produced a concentrated nitrogen fertilizer from liquid pig manure using electrodialysis and reverse osmosis, albeit with significant ammonia volatilization. In scenarios where the required phosphorus concentrations in the effluent are the main drive force, rapid sand filtration provides a simple solution to reduce tertiary phosphorus, primarily associated with solid organic matter [121]. However, rapid sand filtration lags in removal of dissolved phosphorus and does not provide an easily renewable source of phosphorus. In contrast, membrane filtration has gained prominence due to improved performance and cost effectiveness. Tertiary membrane filters, reverse osmosis (RO) systems, and membrane bioreactors (MBRs) have become common in phosphorus recovery, allowing for the collection of both dissolved and solid phosphorus, thereby increasing the phosphorus removal efficiency. These technologies have demonstrated effluent concentrations as low as 0.008 mg P/l for RO systems [122].

Forward osmosis (FO), membrane distillation (MD), and electrodialysis (ED) are three key membrane technologies used for nutrient recovery, each with its own unique advantages. These technologies have been investigated in various combinations, such as hybrid FO–MD systems and hybrid membrane bioreactors (MBR), which show a high efficiency in phosphorus recovery. FO uses a semipermeable membrane to enrich nutrients on the feed side, with the potential for larger phosphate enrichment due to electrostatic repulsion. In MD, heated feed solutions facilitate the conversion of ammonium ions into volatile ammonia, which can be obtained by drawing solution. ED uses cathexis and anion exchange membranes to separate ammonium and phosphate ions, thereby controlling their concentration and regeneration in different chambers [112].

Despite promising results at the laboratory scale, challenges such as membrane fouling, high costs, and the need for the further investigation of salinity control methods remain an obstacle to the widespread practical application of membrane technologies for nutrient recovery [123].

3.4. Chemical Precipitation

The removal of phosphorus from wastewater is often achieved by chemical precipitation using salts of divalent or trivalent metals such as calcium, aluminum, or iron [68,124]. Common precipitants such as lime ($Ca(OH)_2$), alum ($Al_2(SO_4)_3 \cdot 18H_2O$), and ferric chloride ($FeCl_3$) are often used in wastewater treatment plants [100]. The addition of these chemicals initiates the formation of insoluble phosphorus compounds, primarily iron or aluminum phosphate complexes, which are then separated from the liquid phase by settling or filtration [68]. pH adjustments play a critical role in the effectiveness of chemical precipitation. Ferric phosphate precipitates show a minimum solubility in the pH range of 4.0–6.0, while aluminum phosphate precipitates achieve a minimum solubility at pH 5.0–7.0. However, achieving optimal pH levels may require alkalinity adjustments, especially when using calcium-based precipitation methods such as calcium apatite ($Ca_{10}(PO_4)_6(OH)_2$), which requires a high pH (>9.0) for precipitation [104]. Despite the effectiveness of chemical precipitation in removing dissolved phosphorus, the recovery of phosphates from the resulting sludge remains impractical without a tertiary infrastructure for chemical reduction [123]. In addition, the use of chemicals such as lime or alum significantly increases sludge production during primary and secondary treatments, with estimates of up to a 35% increase in the sludge volume after precipitation [104]. Struvite, a crystalline compound composed of magnesium, ammonium, and phosphate ions, spontaneously precipitates in wastewater treatment plants under specific conditions, especially at elevated pH values (>8.0) and turbulent flow. While struvite formation presents operational problems such as pipe clogging, it also offers an opportunity for effective nutrient recovery, particularly phosphorus and nitrogen, by acting as a slow-release fertilizer [123]. Struvite restoration involves dosing the influent with magnesium compounds to induce supersaturation and promote precipitation. Maintaining a stoichiometric molar ratio of magnesium to phosphate is crucial, typically exceeding a ratio of 1:1. However, achieving optimal pH values for struvite precipitation may require aeration or the addition of alkaline agents, thus balancing the cost effectiveness and operational efficiency [104].

3.5. Bioelectrochemical Systems

Bioelectrochemical systems (BESs) represent a breakthrough approach in wastewater treatment, where chemical energy stored in organic matter is directly converted by specific microorganisms into electrical energy. This process not only generates electricity but also provides valuable compounds. At the anode, anaerobic bacteria facilitate the oxidation of organic matter, commonly represented by the oxidation of acetate.

$$CH_3COO^- + 4H_2O \rightarrow 2HCO_3^- + 9H^+ + 8e^- \tag{10}$$

Microorganisms then transfer the generated electrons to the anode, which is connected via the external circuit to the cathode, where reduction reactions take place. BESs are divided into two main types based on the cathodic reaction: microbial fuel cells (MFCs) and microbial electrolysis cells (MECs). MFCs produce electricity by coupling anodic oxidation with electron acceptor reduction, while MECs require electricity to drive a reduction reaction at the cathode, often resulting in the production of hydrogen gas. In MFCs, the cathodic reaction involves the oxygen reduction reaction (ORR), while in MECs, it involves the hydrogen evolution reaction (HER). At near neutral or alkaline pH values, these reactions can be represented as follows [104]:

$$2H_2O + O_2 + 4e^- \rightarrow 4OH^- \tag{11}$$

$$2H_2O + 2e^- \rightarrow H_2 + 2OH^- \tag{12}$$

The potential of a BES as a sustainable wastewater treatment method lies in its ability to produce electricity, regenerate ammonia, and utilize low-quality substrates such as wastewater itself as a source of electrons, thereby reducing the need for external carbon sources.

However, current levels of energy recovery are relatively low, making BESs more suitable for contaminant removal and resource recovery than for energy generation [104].

Biological nitrogen recovery using BESs includes the recovery of ammonia/ammonium from WW. One mechanism involves the biological oxidation of organic matter in the anodic space, which contributes to energy recovery, and at the same time, allows for the regeneration of ammonia/ammonium by migration through the ion exchange membrane to the cathodic compartment with an elevated pH catholyte. The recovery of ammonium from urine combined with the recovery of phosphates through the precipitation of struvites has also been proven. In addition, BESs can promote ammonium oxidation at the cathode, further increasing the nitrogen removal efficiency. Despite the advantages offered by BESs, several challenges remain. The reactor performance is affected by issues such as pH gradients, cation and anion concentration gradients, and potential losses in ion exchange membranes. Recent advances have shown improved performances with high current densities being achieved in certain configurations. However, the scaleup of BESs from laboratory-scale reactors to larger volumes with a reduced internal resistance remains a significant challenge for practical application in larger-scale wastewater treatment. Increasing the current density has been shown to significantly enhance ammonia recovery in microbial fuel cells (MFCs), with approximately 61% of ammonia transport driven by electricity [125]. Microbial fuel cells (MFCs) represent a promising avenue for simultaneous energy production and nutrient recovery. In these systems, microorganisms oxidize organic matter at the anode, releasing electrons that flow through an external circuit to the cathode, where they drive the reduction of nitrate and phosphate ions, thereby facilitating N and P regeneration. In the study, Wu and Modina used microbial electrolysis (MEC) to simultaneously produce hydrogen and regenerate ammonia. By adjusting the pH of the catholyte to 12 through current generation, they achieved impressive results, recovering 96% of ammonia from synthetic wastewater and 79% from real wastewater [126].

In conclusion, a BES has enormous potential for sustainable wastewater treatment, offering simultaneous electricity generation, nutrient recovery, and contaminant removal. Continuous R&D efforts are needed to address the challenges and optimize BES performance for wider application in real-world wastewater treatment scenarios.

3.6. Biological Processes

Microorganisms, including bacteria and fungi, play a key role in nutrient cycling and can be used to recover N and P from waste streams. Some bacteria, called nitrogen-fixing bacteria, can convert atmospheric nitrogen gas (N2) to ammonia (NH_3) through nitrogen fixation, thus offering a sustainable source of nitrogen for agricultural applications [104].

3.6.1. Algae-Based N and P Recovery

Algae, including microalgae and macroalgae, are capable of assimilating and accumulating N and P from wastewater. Microalgae such as *Chlorella* and *Scenedesmus* show high rates of nutrient uptake and are often used in wastewater treatment systems. These microalgae can remove NH_4^+ and NO_3^- from wastewater by biological uptake and simultaneously release oxygen through photosynthesis [127]. In addition, the uptake of luxury phosphorus by microalgae, where phosphorus is stored as polyphosphate in biomass, represents a promising route for obtaining phosphorus. Macroalgae, including water including animal feed and biofuel production [127]. Microalgal growth and further microalgae-based nutrient recovery during wastewater treatment can be affected either positively or negatively by several biotic and abiotic factors. Biotic factors include the presence of bacteria, fungi, viruses, and other microalgae, while abiotic factors include light, temperature, pH, salinity, nutrient, dissolved oxygen concentration, and the presence of toxic compounds [127–130]. Critical to microalgal biomass production is the nitrogen-to-phosphorus molar ratio (N/P) in wastewater. Deviations from optimal ratios, specifically ratios lower than 5 or higher than 30, result in a nitrogen or phosphorus deficiency, respectively, affecting microalgal growth. Various wastewaters, such as those from the pig industry, domestic sources, dairy manure, and anaerobically digested municipal sewage, have been found suitable for microalgal growth when presenting appropriate N:P molar ratios. The nitrogen fixation capability of cyanobacteria enables their survival in low nitrogen environments, offering additional resilience to certain wastewater conditions. However, for optimal growth and nitrogen removal, several key parameters must be carefully managed. The control of CO_2 solubility, impacted by the pH and temperature, is crucial for microalgae photosynthesis. Ammonium volatilization, particularly at an elevated pH and temperature, poses challenges to NH_4^+ removal, necessitating CO_2 enrichment and buffering to mitigate adverse effects on growth and nitrogen removal. Temperature has a significant effect on microalgae growth and nitrogen regeneration, with a specific affinity found for nitrate compared to ammonium. Cyanobacteria strains exhibit different nutrient removal abilities influenced by environmental factors such as light, temperature, and pH. Some strains show effectiveness over a wide range of temperatures, making them suitable candidates for wastewater treatment under various conditions. Light quality and quantity are crucial in microalgae growth and nitrogen removal efficiency. Optimum wavelengths for reproduction and nitrogen removal efficiency vary, with red light recommended for certain species such as *C. vulgaris*. Moreover, wastewater treatment in dark conditions, especially through combined cultivation systems, represents an economically and energetically efficient approach [131].

Valchev and Ribarova [132] analyzed the microalgae-based nutrient recovery technologies for tertiary treatment of municipal wastewater. They are considered promising green technologies with many economic and environmental benefits. In the field of municipal wastewater treatment, algae-based technologies are being investigated for multiple applications, such as for the removal and recovery of the macronutrients N and P, decomposition of organic pollution, elimination of pathogenic microorganisms, and removal of micropollutants and heavy metals. Microalgae are most often used to remove nitrogen and phosphorus. The authors of [132] focused on microalgae-based technologies for tertiary treatment aimed at removing P and N after conventional secondary biological treatments at municipal wastewater treatment plants (MWTPs). The advantages of tertiary cleaning using algae are evidenced by the elimination of phosphorus precipitants, energy savings for the provision of recirculation ponds for biological nitrogen removal, and an improvement in the quality of the excess biomass created and its further application through the safer use of N and P. The use of algae also increases the quality of treated WW by reducing its acidity and by increasing the content of dissolved oxygen. However, despite these advantages, as well as extensive research and development efforts, these algal technologies have not been successfully applied to N and P removal and recovery on an operational scale in WWTPs. Based on the literature review, the authors tried to analyze and identify the main causes of this condition. The authors of [132] discuss the main aspects of the reliability and readiness level of the technologies being developed.

3.6.2. Recovery of N and P Based on Macrophytes

Another biological component, macrophytes, such as aquatic plants including duckweed (*Lemnaceae*), water lettuce (*Pistia stratiotes*), and water hyacinth (*Eichhornia crassipes*), play an important role in the removal and recycling of nutrients in aquatic ecosystems. These plants can absorb and accumulate N and P from water through their root systems, thus providing a natural and sustainable way to renew nutrients. Duckweed has proven its effectiveness in removing NH_4^+ and NO_3^- from wastewater, while the collected biomass serves as a potential raw material to produce bioethanol or other valuable chemicals. In addition, emergent macrophytes planted in constructed wetlands can enhance nutrient removal through physical uptake, precipitation, and biological assimilation processes [122].

The authors consider the fundamental challenges of further research for all types of reactors to be the identification of optimal algal strains, the determination of technological parameters, and overcoming problems related to seasonal variability of WW quality and problems with biomass collection.

4. Prospects in Nitrogen and Phosphorous Removal and Recovery

Considerable developments have been achieved in the last decades around nutrient elimination from wastewater, but a lot of challenges are still present. Many studies confirmed the feasibility and performance of conventional methods for the simultaneous elimination of organics and nutrients. Conventional processes were modified based on the configuration of the reactors, sequences of the zones, pollution loads, retention time, and optimization of operational conditions. Modifications of the metabolism and microbiology involved in the elimination of nitrogen and phosphorous were also implemented. The up-scaling of the processes described in the scientific literature will require intensive studies focusing on the development of low cost and efficient removal strategies for nitrogen and phosphorous from wastewater using a combination of aerobic, anoxic, and anaerobic processes or more significant modifications of metabolic pathways from a microbiological aspect. To understand, in detail, the role of microbial communities and biofilm development together with selection of approaches to optimize the biomass density is crucial. These parameters determine the practical choice of the system for nutrient elimination. The relationships among various groups of microorganisms within the treatment community should be explored to understand useful interactions between microorganisms to obtain the required levels of contaminant elimination. Partial nitritation and anammox for nutrient elimination is an approach with great potential, because it decreases the energy demand for aeration. A major obstacle is the slow development of anammox bacteria at lower temperatures. The system is prone to inhibition in large, open-air systems in variable weather situations. The produced nitrite must be stable enough to support the process. The anammox process, besides its benefits, also has an important limitation related to quite high concentrations of nitrate in the effluent. This usually requires an additional step of post-denitrification to accomplish the legislative limit. As a result, a higher price of treatment is expected. An elevated C:N ratio and decrease in temperature are the major challenges when the anammox method for treating wastewater is considered. In the case of the organic matter present, heterotrophic denitrifying bacteria compete with anammox bacteria, resulting in a poor ammonium removal efficacy. Various organics, such as methanol, also inhibit anammox processes. Numerous challenges related to microalgae-based methods must be also solved for the large-scale operation. This approach needs large land regions for establishing appropriate water bodies/cultivation basins. They are significantly affected by weather conditions and the pollution load. At the same time, algae harvesting options must be considered as well as the final phosphorous concentration procedure and procedure yield. All of this addresses the profitability of an approach to provide consistent nutrient elimination. No matter what technology for removing of macronutrients from wastewater is considered, not only the removal efficiency but also the P and N recovery rate is important. As a part of circular economy, a paradigm shift is needed. Phosphorous and nitrogen from wastewater are recognized as valuable raw materials to produce fertilizers as well as other useful products, and thus, should be retained in the economic cycle. The fertilizer production market uses 80% of natural, non-renewable phosphorus resources in the form of phosphate rock. The depletion of these increases the need for other alternatives, including wastewater and sewage sludge. Wastewater is a resource with a relatively low P concentration and requires well-thought-out and well-designed treatment to recover the highest possible amount of phosphorus. A compressive evaluation of the combined methods is crucial for future research in this area.

5. Conclusions

The discharge of nutrient-rich wastewater poses significant challenges to aquatic ecosystems, leading to eutrophication. The effective removal of nitrogen and phosphorus is essential to meet stringent discharge standards and prevent environmental degradation. Additionally, nutrient recovery plays a crucial role in realizing a circular economy and conserving limited resources. This article was prompted by a draft revision of the "Urban Wastewater Treatment Directive", highlighting the importance of selecting appropriate nutrient removal technologies. The choice of technology depends on various factors, including wastewater characteristics and treatment goals. While previous treatment efforts have yielded positive results, new ambitious goals necessitate a comprehensive approach to protecting water ecosystems and human health. This paper provides operators and designers with an overview of traditional and advanced processes for nutrient removal from municipal wastewater, along with discussions on compliance with stricter effluent standards. There are various options available to intensify existing treatment plants, such as optimizing nitrification processes and implementing cyclical nutrient removal processes. The discovery of new microbial communities offers promising technologies for efficient nitrogen removal, potentially leading to a higher throughput and reduced energy consumption. Chemical precipitation is crucial for consistently maintaining phosphorus levels below regulatory limits. Overall, the effective management of nitrogen and phosphorus during wastewater treatment is vital for safeguarding water resources and promoting environmental sustainability.

Many scientific publications can be found in the literature that draw conclusions or make claims about the capabilities of specific technologies in reaching low nutrient concentrations in the effluents. At the same time, they imply various techniques for phosphorous recovery. However, many of these publications are useful, but the level of their technical performance should be sometimes viewed with a great degree of caution. They are not supported by data from pilot- and real-plant design and detailed operational information, and there is a lack of statistical analyses of data from long-term operating periods. Data from the lab-scale studies without any long-term statistical monitoring and evaluation could be misleading to wastewater treatment plant management personnel making infrastructural decisions to comply with new nutrient permit limits.

Author Contributions: Conceptualization, J.D., A.Ž.G. and A.K.; methodology, J.D., A.Ž.G. and P.G.; software, P.G.; validation, J.D., A.Ž.G. and P.G.; formal analysis, J.D., A.Ž.G., P.G. and A.K.; investigation, A.K., P.G. and N.Š.; resources, J.D.; data curation, J.D.; writing—original draft preparation, J.D., A.Ž.G., P.G. and A.K.; writing— review and editing, J.D., A.Ž.G. and P.G.; visualization, P.G. and N.Š.; supervision, J.D.; project administration, J.D. and A.K.; funding acquisition, J.D. and A.Ž.G. All authors have read and agreed to the published version of the manuscript.

Funding: This work was supported by the Slovenian Research Agency ARRS (Research Program Chemical Engineering P2-0191).

Conflicts of Interest: The authors declare no conflicts of interest.

Abbreviations

ACZ	anoxic contact zone
ANOX	anoxic zone
ANAMMOX	anaerobic ammonium oxidation bacteria
AO	anaerobic/oxic
AOA	ammonia oxidizing archaea
AOB	ammonium oxidizing bacteria
ATP	adenosine triphosphate
BABE	bioaugmentation batch enhanced treatment
BNR	biological nutrient removal
BOD(s)	biological oxygen demand(s)
C	carbon
CAS	conventional activated sludge
CNR	cyclical nitrogen removal
COD	chemical oxygen demand
COMAMMOX	complete ammonia oxidation bacteria
CSTR	continuous stirred-tank reactor
DAMOs	anaerobic methane oxidation denitrifying microorganisms
DBSS	digested biological sewage sludge
DO	dissolved oxygen

DPAO(s)	denitrifying polyphosphate-accumulating microorganism(s)
E	efficiency
EBPR	enhanced biological phosphorous removal
ED	electrodialysis
EU	European Union
FEAMMOX	iron reduction bacteria
FWR	food waste-recycling wastewater
HNAD	heterotrophic nitrification and aerobic denitrification
HRT	hydraulic retention time
IR	internal recirculation
LE	Ludzack–Ettinger system
MBR	membrane bioreactor
MD	membrane distillation
MF	microfiltration
MFCs	microbial fuel cells
MLE	modified Ludzack–Ettinger (system)
MLSSs	mixed liquor suspended solids
N	nitrogen
O	oxic zone
P	phosphorous
PAO(s)	polyphosphate-accumulating microorganism(s)
PE	population equivalent
PHA	polyhydroxyalkanoates
PPK	polyphosphate kinase
R	recirculated liquid
R	regenerator
RO	reverse osmosis
RS	return sludge
SAFF	Submerged Aerobic Fixed Film Reactor
SBR	sequential batch reactor
SHARON	single reactor for high activity ammonia removal over nitrite
SND	simultaneous nitrification and denitrification process
ST	sedimentation tank
SRT	solid retention time
SVI	sludge volume index
SWOs	Stormwater Overflows
TBSS	thickened biological sewage sludge
ThAlMBR	Thermophilic Alternate Membrane Biological Reactor
TKN	total kjeldahl nitrogen
TN	total nitrogen
TP	total phosphorous
TSSs	total suspended solids
UCT	University of Cape Town
UWWTD	Urban Wastewater Treatment Directive
VFA	volatile fatty acids
VIP	the Virginia Plant Initiative
WS	waste sludge
WW	wastewater
WWTP	wastewater treatment plant

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