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Simultaneous Removal of Organic Matter and Nitrogen in Constructed Wetlands. Critical Review.

Eliminación simultánea de materia orgánica e nitróxeno en humedais construidos. Revisión crítica.

Eliminación simultánea de materia orgánica y nitrógeno en humedales construidos. Revisión crítica.

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Abstract

Spurred by the needs to find a better treatment method for wastewater, a method that is cheap, less complex when compared to conventional methods, and more importantly, that is climate friendly and has low cost of maintenance and operations, the use of constructed wetlands that is well designed, optimally operated, and of the best type of configuration has been proven by environmental scientists to meet these objectives. Constructed wetlands have proven to be very useful for the simultaneous elimination of nitrogen and organic matter as well as the removal of solids and pathogens contained in wastewater; it can achieve an efficiency as high as 70-94% NH_4^+removal, 70-96% N removal, 80-95% BOD and 73-96% COD removal.

In this study, a profound review is made of the broad treatment process of wastewater from the primary treatment to the tertiary treatment, with the use of the conventional methods, analysis of constructed wetlands, its components, types and functions of each components, the various types of macrophytes and in their best operating environment, as well as the types of micro-organisms present. Also, a special focus is made about the characterization of wastewater, the parameters that are used, the impact of fairly treated or when wastewater is not treated at all, on humans and the environment. Analysis of the physico-chemical and biological processes involved in the removal mechanisms of both nitrogen and organic matter and, of course, how environmental factors and operating parameters influence the effectiveness of the treatment process and lastly, analysis of promising methods, like CANON, ANAMMOX, or SHARON, that are being investigated with the aim of implementing them in the future designs of constructed wetlands.

Keywords:

Wetlands, Constructed Wetlands, Macrophytes, Nitrification, Denitrification, Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD).

Resumo

Existe a necesidade de atopar métodos de xestión das augas residuais, máis sinxelos que os métodos convencionais, respectuosos co medio ambiente e con baixos custes de mantenimiento e operación. Entre estes métodos alternativos atópanse os humidais construidos que, deseñados e operados de maneira óptima, dan resposta a estes obxectivos. Se ten demostrado que os humidais construidos permiten a eliminación simultánea de nitróxeno e materia orgánica, así como de sólidos e patóxenos, en augas residuais. Os resultados amosan eficacias de eliminación entre 70-94% de NH_4^+, 70-96% de N, 80-95% de DBO e 73-97% de DQO.

Neste traballo faise unha revisión do proceso de tratamento de augas residuais empregando os métodos convencionais, dende o tratamento primario até o terciario, para posteriormente centrarse na análise dos humedais cosntruidos, tipos e funcionalidades dos seus componentes, tipos de macrófitas empregadas e as mellores condicións ambientais, así coma microorganismos presentes. Préstase especial atención á caracterización das augas residuais, os parámetros que se estudan e o impacto das augas non tratadas ou moderadamente tratadas sobre os humanos e o medio ambiente. Analízanse os procesos físico-químicos e biolóxicos implicados nos mecanismos de eliminación de nitróxeno e materia orgánica e, por suposto, a maneira na que os factores medioambientais e os parámetros operacionais inflúen na eficacia do proceso. Por último, faise referencia a varios métodos prometedores, CANON, ANAMMOX e SHARON, que están sendo investigados para a súa implementación en futuros deseños de humedais construidos.

Palabras chave:

Humedais, Humedais Construidos, Macrofitos, Nitrificación, Desnitrificación, Demanda Biolóxica de Osíxeno (DBO), Demanda Química de Osíxeno (DQO).

Resumen

En la búsqueda de mejores métodos de gestión de aguas residuales, más simples que los métodos convencionales, respetuosos con el medio ambiente y con costes de mantenimiento y operación bajos, se ha encontrado que los humedales construidos, diseñados, configurados y manejados de manera óptima, dan respuesta a estos objetivos. Se ha demostrado que los humedales construidos permiten la eliminación simultánea de nitrógeno y materia orgánica, así como de sólidos y patógenos, en aguas residuales; los resultados muestran eficiencias de eliminación entre 70-94% de NH_4^+, 70-96% de N, 80-95% de DBO y 73-97% de DQO.

En este trabajo se hace una revisión del proceso de tratamiento de aguas residuales empleando los métodos convencionales, desde el tratamiento primario hasta el terciario, para posteriormente centrarse en el análisis de los humedales construidos, tipos y funcionalidades de sus distintos componentes, tipos de macrofitos empleados y sus mejores condiciones ambientales, así como microorganismos presentes. Se presta una particular atención a la caracterización de las aguas residuales, los parámetros que se estudian y el impacto de las aguas no tratadas o moderadamente tratadas sobre los humanos y el medio ambiente. Se analizan los procesos físico-químicos y biológicos implicados en los mecanismos de eliminación de nitrógeno y materia orgánica y, por supuesto, el modo en que los factores medioambientales y los parámetros operacionales influyen en la eficacia del proceso. Por último, se mencionan varios métodos prometedores, CANON, ANAMMOX y SHARON, que están siendo investigados para su implementación en futuros diseños de humedales construidos.

Palabras clave:

Humedales, Humedales Construidos, Macrofitos, Nitrificación, Desnitrificación, Demanda Biológica de Oxígeno (DBO), Demanda Química de Oxígeno (DQO)

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List of acronyms

BOD: Biological oxygen demand

COD: Chemical oxygen demand

CW: Constructed wetland

DO: Dissolved oxygen

FSCW: Free surface constructed wetlands

HF-SSCW: Horizontal flow sub-surface constructed wetlands

HLR: Hydraulic loading rate

HRT: Hydraulic retention time

HSWW: High strength wastewater

LSWW: Low strength wastewater

MSWW: Medium strength wastewater

SSCW: Sub-surface constructed wetlands

TDS: Total dissolved solids

TN: Total nitrogen

TOC: Total organic carbon

TSS: Total Suspended solids

VF-SSCW: Vertical flow sub-surface constructed wetlands

VSS: Volatile suspended solids

WW: Wastewater

1. INTRODUCTION

Water has always been very important to human; in fact, water has been a determining factor in choosing a settling place for man starting from the stone era; it determines among other factors the survival of human in an environment early man knew little or nothing about. In fact, it would have been really difficult for life to exist if there is no water.

Human's use of water is immensely complex, diverse, and span through almost all the activities that humans engage in, making water to be indispensable. When water is used; be it in a domestic setting, industrial, hospital, social parks, agricultural, etc., its composition changes, and when it is returned to the environment it becomes dirty water or better still wastewater (WW). WW is usually discharged to gutters; (open drainage), close drainage, lakes, rivers, sea, or even grass, flowers, uncompleted buildings, roofs of some houses, etc. it all depends on how effective municipals orders, policies on wastewater are and its enforcement, the social awareness of the effects; which most often are negative, of wastewater discharge, and the level of development of the cities, etc.

Due to rapid urbanization, industrialization, rapid standard of living, rapid change in lifestyle, water consumption has sharply increased so does the volume of wastewater discharge into the environment. The problem comes in when one bears in mind that most wastewater discharge are either fairly treated or not treated at all, causing a lot of diseases and environment problems.

To combat this problem, various conventional methods have been designed, methods like chemical coagulation, flocculation, adsorption, oxidation, membrane bioreactor, etc. Although all these methods have all been widely applied, they do have some drawbacks, like adsorbents being difficult to regenerate, chemical coagulants could lead to further undesired reactions in water bodies and possibly further pollution, also coagulants generating a lot of sludge. Chemical oxidation if not well controlled could have adverse effects on aquatic life and even on humans. Other conventional methods include activated sludge, biological trickling filters, and rotating biological contactors. Both biological trickling filters and rotating biological contactors are very sensitive to temperature and their removal rate of organic matter is low while activated sludge on the other hand requires a lot of energy to operate the system appurtenances like blowers, pumps, etc. Generally conventional methods are very expensive to maintain and generate large volumes of greenhouse gases that compound the problem of climate change.

Health and environmental institutions with noble intentions of curtailing the adverse effects of wastewater, keep churning out policies that would protect the general health of the populace and safe-guard the environment, sometimes modifying policies that are already in place or in extreme cases sponsoring new bill(s) to be passed by the parliament into law(s). Environmental laws that are being passed, modified or made to replace existing ones are becoming stricter, as they aim to limit the quantity of contaminants, either as a single specie or as a group, that could be discharge or emitted to the environment. These policies place a lot of pressure on increasing the efficiency of conventional methods to treat wastewater. The consequence is that conventional methods become more expensive, emit more greenhouse gases and could in some cases lead to break down of the plants.

A way out of this dilemma is the use of constructed wetlands (CW). It is one of the unconventional treatment methods that, provided it is well operated and well designed, offers better wastewater treatment efficiency. Although it has its drawbacks, it is less sophisticated in terms of maintenance and operation and is a low-cost treatment method. This paper will make a critical analysis of the followings:

- A detailed analysis of the characteristics of WW; physical chemical and biological parameters, its strength, types, the sources of WW, adverse impacts of the discharge of WW and the general process of WW treatment.
- A profound analysis of constructed wetland (CW), its history, types, the role its components like macrophytes plants, microbes etc., play in the physio-chemical-biological processes in the removal mechanism of nitrogen and organic materials and also the types of microbes present in CW. An analysis of the biological, physical and chemical mechanisms of the removal of Nitrogen and organic material in different conditions; anaerobic, aerobic and anoxic, the factors which influence the mechanisms and the optimum condition, the classical routes and the innovative ones.
- The effect of environmental factors like temperature, dissolved oxygen, pH, etc. and operational conditions like hydraulic and retention loading time, influent feeding mode, external organic source, recirculation, etc. on the removal process of nitrogen and organic material.
- And useful considerations that could help in raising the efficiency of treating WW using CW now and some in future research.

2. WASTEWATER

2.1. Types of wastewater

Basically, there are three types of wastewater: Domestic, Industrial and Stormwater run-off wastewater. Stormwater is water from rainfall and sometimes from melting ice which has not been absorbed by the ground becoming surface run-off. Storm water flows from rooftops of buildings, lawns, streets, driveways, parking lots, construction sites, municipal storage yards, etc. Storm water run-off carries many pollutants which vary from place to place. The pollutants mostly found are: Oil and grease, Garbage, gasoline from automobiles, sediments from construction sites, metal flakes, road salt (halite), which is usually common during winter, pesticides. Other pollutants found in storm water run-off also include Herbicides, heavy metals from roof shingles, pet waste, leaves, grass clippings, bacteria, illicit discharges from paints, cleaning solution products, used motor oil, etc.

Industrial wastewater on the other hand contains pollutants that are characterized by the bio-chemical and physical activities of the industries. They are mostly aqueous and organic discharges of varying concentration. Sometimes they may include toxic pollutants as well as other components that are harmful to the environment and more worrisome, are difficult to degrade either through biological or chemical operations. Consequently, the treatment of some industrial wastewater could be specially complicated which could need specially advanced and complex methods.

Domestic wastewater: This is wastewater derived due to human activities in households like kitchen, toilets, bath, dish washing machine, laundry, spas, etc. It contains 99% water as a result it contains very little concentrations of pollutants when compared to industrial wastewater. Basically, it can be grouped into three types: Black water, Grey and Yellow water.

Black water: It is the wastewater from the toilet, kitchen and the dish wash. The composition includes poop, toilet paper, cleaning liquid, urine, water, cleansing water, etc. It is termed blackwater because of its high potential of being contaminated by pathogens and grease [2a].

Greywater: It is wastewater from bathtubs, sinks, washing machine, showers; it’s any wastewater that does not originates from the toilet. It is less contaminating when compared with Blackwater.

Yellow water: This results from wastewater that has been carefully designed to originates from urine, that explains the origin of the name.

2.2. Characterization of wastewater

The source of wastewater being studied definitely determines its characteristics, be it domestic, industrial or storm run-off. Also, to design an effective treatment plant for wastewater it is extremely important to have enough information of the wastewater, for instance, the wastewater from a brewery industry is definitely different in concentration and composition from that of a dairy farmland.

The characteristics of the effluent wastewater is wholly influenced by that of the influent wastewater. The vital information of a wastewater comes from its characterization using the following parameters:

A) PHYSICAL PARAMETERS:

1) **ELECTRICAL CONDUCTIVITY**: It's a parameter that indicates the ability of wastewater; liquid generally, to conduct electricity. This ability is indicated by the concentration of the conductive ions present in the wastewater. When chemicals, biological compounds as well as salts dissolve in water, they dissociate forming positive ions, cations, and negative ions, Anions. The cations commonly found in wastewater are potassium ions, magnesium, and odium ions while anions include chlorides, sulphates, sulfides, carbonates, etc. [3a]. It's measured at a standard temperature of 25°C so as to ensure comparison of reading under different climatic conditions [4a,5a].

As pure water has an extremely low concentrations of both cations and anions, it has an extremely low conductance, as a result, a significant change in the electrical conductance of wastewater indicates the presence of a contaminant or absence of some components. Electrical conductivity is related to Total solids of wastewater *(Table 1)*.

Table 1. *Values of the conductivity of various types of water*

Types of water	Conductance
Pure distilled and de-ionised water	*0,05 µS/cm*
Seawater	*50 mS/cm*
Drinking water	*200-800 µS/cm*
Rain or Snow water	*2100 µS/cm*

2) **SOLIDS:** Generally, solids analysis in wastewater is essential in the control of physical and biological processes in wastewater treatment and more importantly in assessing the compliance with standards set by various regulatory agencies. Although wastewater has an approximately 99,9% liquid composition and 0,1% solid composition, the solid composition sometimes gives an interesting information. The amount of solids in wastewater is sometimes used to indicate the strength of the wastewater, the more the solid composition the more the strength of the wastewater. The total dissolved solids, TDS, is important for drinking water and irrigation, the total suspended solids, TSS, is used as a standard for water that is safe for consumption while Volatile suspended solids, VSS, gives an indication of the organic matter present in the wastewater.

I. **Total Solids (TS):** Total solids is the summation of total dissolved solids and total suspended solids present in wastewater or any liquid that is under investigation, it is measured in mg/l. Solids may be classified as organic or inorganic, filterable or non-filterable. Filterable solids could be settleable or non-settleable. Total solids present in wastewater gives an indication of the effect of run-off from construction site, agricultural practice, logging activities and of course sewage treatment plant effluents. It is determined in the laboratory by heating a measured volume of a sample in a crucible dish placed in a drying oven at 103-105°C.

II. **Suspended Solids (SS):** It is the small solid particles that remain as suspension in wastewater, it's also an indicator of water quality as it has a direct effect of wastewater treatment [6a,7a]. Although it has the same abbreviation as Settleable solids, the two solids are completely different, it's also measured in mg/l. It is usually determined in the laboratory by filtering the sample on a previously weighed glass-fibre filter and drying the filter at 105°C. Later on, the filter is then cooled to room temperature and weighed.

III. **Dissolved Solids (DS):** It's sometimes referred to as the non-filterable solids, the solids that pass through a 2,0 micro pore size filter when the sample is filtered. The filtrate is dried and weighed. The dissolved solids of water meant for public consumption should be less than 500mg/l [8a].

3) **TURBIDITY**: Turbidity is an optical property that is used to describe the haziness or cloudiness of water. It is due to the presence of suspended materials like clay, silt, organic and inorganic materials that are of very small but of different diameters which disperse electromagnetic radiation of Infrared and Visible light region and it's also due to the presence of dissolved coloured organic compounds which absorb these radiations [2]. In wastewater treatment, organic suspended material is usually observed [3]. Turbidity is a parameter used in measuring water quality; the more turbid a wastewater sample is, the lower is the quality [9a]. Turbidity is measured in nephelometric turbidity unit (NTU), for water meant for huma consumption its turbidity should be less than 1,0 [9a].

4) **TEMPERATURE:** Temperature is a parameter that directly affects biological and chemical reactions so much so that the temperature of a wastewater treatment plant could indicates the degree of metabolism. Generally, the temperature of wastewater is between the range of 10 and 20°C but it must be stressed that the mean temperature varies depending on the source of the wastewater being treated [10a].Temperature also affects the quantity of dissolved oxygen in wastewater as oxygen is more soluble in cold water than in warm and since the optimum temperature of bacteria activity which is in range of 25-35°C, it means the temperature of the treatment plant needs to be adjusted without compromising the availability of dissolved oxygen.

5) **COLOUR:** The colour of an untreated wastewater is normally brownish grey but as time passes by it changes its colour to deep grey and on the long run to black [1a].

B. CHEMICAL PARAMETERS:

1) **Alkalinity:** Alkalinity is a parameter that is used to measure the capability of water to neutralize acids. It is due to the presence of some compounds in water like hydroxide, carbonate, bicarbonate of sodium, potassium, ammonia and of magnesium. The activity of most microbes in wastewater treatment operates at an optimum pH of 7,0-7,2 (in some textbook 7,0-7,4), but the problem is that the microbial operations generate acid which lowers the pH. Consequently, it is imperative to maintain a pH for optimum bio-activities. This is achieved by the presence of sufficient alkalinity which neutralizes the acid produced. Alkalinity is measured by titrating a given sample with a

standard acid until a pH of 4,5 is attained [11a,12a]. Total alkalinity is calculated as the sum of the concentrations of $[OH^-]$, $2[CO_3^{2-}]$ and $[HCO_3^-]$, it is measured in mg of $CaCO_3$/l.

2) **pH:** It is the negative logarithmic activity of the hydrogen ion in wastewater solution. As explained in the previous paragraph, metabolic activities of bacteria are pH sensitive, something which makes pH as a very vital tool in monitoring the efficiency of the wastewater operations. It notifies the point which alkalinity stops, and acidity begins, it tells the alkalinity or acidity of a wastewater solution, but it does not tell the concentration not the composition of the alkalinity of the solution [12a].

3) **Dissolved Oxygen (DO):** In biological treatment, it is the relative measure of the molecular oxygen dissolved in wastewater that is available to sustain the life of micro-organisms present including bacteria. Water solubility of oxygen is 40mg/l at 25°C at 1 bar, its solubility depends on partial pressure, temperature; the solubility decreases at higher temperature, and the presence of dissolved solids [13a]. Micro-organisms uses oxygen to oxidize wastes for growth while releasing energy in the atmosphere, consequently, it's imperative to control the dissolved oxygen during waste treatment especially during the secondary stage [14a]. The DO concentration is vital in monitoring the process of nitrification as well as denitrification at various points of the treatment process. It is measured using an optical and electro-chemical sensor.

4) **Biological Oxygen Demand (BOD):** Essentially, BOD is a measure of the dissolved oxygen consumed by bacteria and other micro-organisms to decompose organic matter in an aerobic condition and at a specified temperature. The organic matter present in the wastewater; carbohydrate, protein, etc. are fed upon by the micro-organisms in wastewater, predominantly bacteria, in the presence of oxygen to give more biomass of the microbes, energy and carbon dioxide according to this equation:

$$\begin{aligned} &\text{organic matter} + \text{nutrient} + O_2 + \text{micro-organisms} \rightarrow \\ &\quad \rightarrow CO_2 + H_2O + \text{energy} + \text{more bio-mass cells} \end{aligned} \tag{1}$$

BOD is usually carried out in a period of 5 days, which explains why BOD is commonly denoted as BOD_5, and it is commonly carried out at 20°C. BOD is affected by any factor that would affect dissolved oxygen, factors like pH, temperature, types of micro-organisms, salinity, atmospheric pressure, etc [7a]. A high BOD means a high composition of easily degradable organic matter is present in the wastewater while a low BOD indicates a low concentration of organic matter possibly difficult to decompose is present [15a].

BOD, precisely BOD_5, is a parameter that is widely used in wastewater treatment to do the followings:

i) To determine the approximate amount of dissolved oxygen necessary to stabilize the organic materials.
ii) Useful in determining the size of wastewater plant to be designed.
iii) To monitor the bio-chemical processes
iv) To determine the efficiency of the overall treatment process [4].

BOD is usually measured using the Meter and Probe methods, Winkler method, etc. in mg/l. Examples of values of BOD of different types of wastewater are given in *Table 2*.

Table 2. *BOD values of different sources of wastewater [4a]*

Types of water	**BOD values (mg/l)**
Pristine river	*≤ 1,0*
Moderately polluted river	*2-8*
Well treated polluted municipal sewage	*20*
Untreated sewage	*≥ 200*
Influent wastewater	*≈ 250*
Effluent wastewater	*≤ 30*
Industrial wastewater	*30000*

5) **Chemical Oxygen Demand (COD):** COD is a measure of the amount of oxygen required to chemically oxidize soluble and particulate organic matter present in wastewater in an acidic condition, it is expressed as the mass of oxygen consumed per litre of solution (mg/l). As it is the case with BOD, a high COD indicates a very high composition of both degradable and difficult to degrade organic content. The test is often carried out with a very strong oxidizing agent like potassium dichromate, potassium iodate, potassium permanganate, under acidic condition.

During the analysis, both organic and inorganic materials are chemically oxidized, making the COD values higher than the BOD. COD offers numerous advantages over the BOD, like:

i) COD requires between 2-3 hours unlike BOD that takes 5 days.
ii) It can be used to test wastewater that contains toxic materials, too toxic for BOD [7a].
iii) All organic materials are oxidized in COD unlike BOD where only degradable material are consumed.

Measurement of COD is carried out using Open Reflux method, Closed Reflux-Titrimetric method, Closed Reflux-Colorimetric method, etc. Although COD and BOD_5 are both independent tests, it's been observed that there is an empirical relationship between the two parameters, a relationship known as bio-degradability index. The ratio of COD: BOD_5 is always constant, for example the bio-degradability index for food processing wastewater, COD: BOD_5 = 2,1 while that of textile, COD: BOD_5 ≈ 5,1 [7a].

6) **Total Organic Carbon (TOC):** It refers to the levels of carbon in wastewater. The TOC value of wastewater indicates the level of organic contamination it has. Since wastewater contains both natural and synthetic organic matter, like amine urea, faecal matter, etc which are organic and pesticides, detergents, fertilizers, etc, which are synthetic, the TOC value indicates the condition

and state of contamination of the wastewater [16a]. It is usually measured in µg/l with TOC analysers which could be Combustion analyzers or Wet Chemical.

7) **Total Nitrogen (TN):** It is an analysis undertaken to quantify the total amount of nitrogen contained in wastewater. It is the sum of organic-nitrogen, organic- bonded nitrogen, nitrate-nitrogen and nitrite nitrogen. Analysis is done for each of this form in which nitrogen exist, the total gives TN. During the bio-chemical processes of nitrogen conversion *(Fig. 1)*, when ammonia, nitrite is at minimum level, the nitrate concentration reaches its peak, the wastewater treatment is often said to be well nitrified [17a]. It's measured in mg/l the Chemscan analysis methods or Laboratory analysis methods.

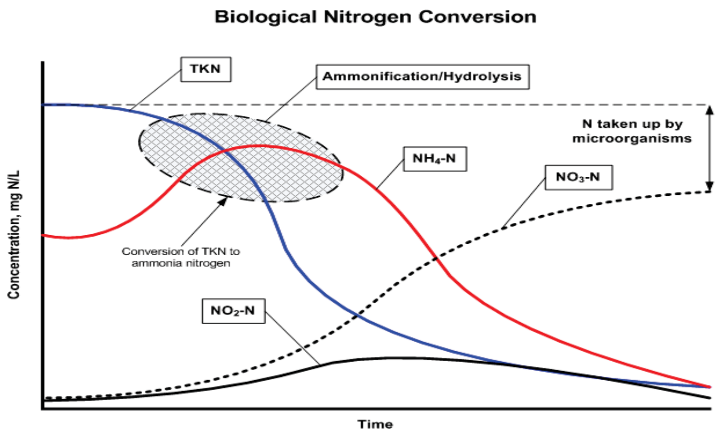


Figure 1. *Biological nitrogen conversion and consumption*

8) **Total Phosphorus (TP):** Phosphorus occurs in wastewater as phosphates, which could orthophosphates, condensed phosphates and organic phosphates. TP is the analysis of all forms of compounds containing phosphorus in wastewater, mostly carried out in an acid condition for preservation then followed by acid digestion. It's commonly measured using the Ion-exchange chromatography method. Municipal wastewater may contain 5-20mg/l, most of which is inorganic [18a].

9) **Fat, Oil and Grease (FOG):** In this test, a specified compound is not being identified and quantified rather a group of compounds that share similar characteristics like solubility in the same solvent is being analysed. Consequently, oil and grease test is actually a test for any compounds extracted by the solvent used. It means oil, grease as well as materials like certain organic dyes, sulfur compounds, chlorophyl fall into this group. Firstly, the sample is acidified, n-hexane is used as an extraction solvent if sample is a contaminated water, petroleum ether is used for natural and treated water. Trichlorofluoromethane is suitable for types of sample but for environmental concern, a mixture of Hexane and methyl tertbutyl ether; 80:20 is now used as an extraction solvent. Partition Infrared method, Soxhlet method, liquid/liquid phase and Solid-phase gravimetric method are used [19a,20a,21a].

C. BIOLOGICAL PARAMETERS:

1) **Total coliforms:** It is a characterization test that encompasses faecal coliforms as well as micro-organisms in soil, it is commonly used as an indicator of water pollution.

2) **Faecal coliforms:** It is an indicator of water pollution with faecal coliforms, the lead indicator is Escherichia coli, a specie of bacteria.

3) **Helminth:** It's an analysis for the helminth eggs in wastewater.

2.3. Strength of wastewater

Most tests carried-out on wastewater used to be done with the sole aim of obtaining the composition of the organic matter but with an objective of classifying the strength of the wastewater. Today, there is no standard definition for classifying wastewater according to their strength, but these tests are usually carried-out to know the organic content, BOD, COD, TOC, TSS and FOG. The concentration of BOD, SS and FOG for domestic wastewater are not usually high when compared to other types of wastewater, an example is given in *Table 3* below:

Table 3. *Parameters of different sources of wastewater [22a]*

Wastewater	TSS (mg/l)	BOD_5 (mg/l)	COD (mg/l)	FOG (mg/l)	TN (mg/l)
Municipal WW	350-1200	100-350	210-740	30-100	20-80
Household WW	252-3320	112-1101	139-1650	16-134	44-189
Storm run-off water	112-1894	12-19	82-178	<1-7	3,5

Most domestic WW has a strength of BOD=100-350mg/l, TSS= 100-350mg/l and when the strength of a wastewater is a lot higher than these values, the WW is most often said to be of higher strength wastewater (HSWW). Industrial wastewaters are usually of high strength, in fact they are even classified as low, medium and high strength:

A) **Low HSWW**: Wastewater emanating from toilet flushing of non-domestic sources with BOD_5 and TSS levels approximately 2 or 3 times that of domestic.

B) **Medium HSWW**: This time around the BOD, TSS and FOG levels around 4-10 times the domestic level. They are WW from kitchen of commercial centres.

C) **High HSWW**: Industrial sources of WW with high BOD and TSS levels and sometimes FOG, that are more than 10 times that of the domestic level.

When high strength WW is treated more attention is placed on the organic carbon content and the FOG level [23a,24a] as well as the bio-degradability of the WW, as both play important role during the biological process. WW is said to be biodegradable when the BOD/COD ≥ 0,5.

2.4. Why treat wastewater?

WW contains many components that have an adverse effect on humans and the environment if it's returned to the environment untreated. Basically, the following are the benefits of treating WW:

- **EFFECTS ON HUMAN**: When WW is treated and released into the environment, the health risks that would have occurred are reduced to the barest minimum thereby making the environment habitably safe. Health risks like the decaying organic matter competing with fish and other wildlife population for dissolved oxygen, leading to oxygen depletion. Untreated WW can contaminate crops and drinking water with its negative consequence on human life and the eco-system. This on the long run save money for other pressing societal needs.

  Pathogenic microbes like bacteria, virus, protozoa, could cause pandemic diseases on human if proper monitoring is not carried out on its treatment and of course its discharge into the environment. Coronavirus is a classic example.
  For heavy industries that consume enormous volume of water and consequently produces a lot of volume of WW that contains heavy metals that cannot be discharged into the sewers, having on-site treatment plant reduces the cost of transportation of transporting WW to the municipal treatment plant as the volume of WW is reduced.

- **EFFECTS ON THE ENVIRONMENT**: Excessive nutrients from untreated WW released into water bodies can cause excessive plant growth, alter habitat and could even lead to the extinction of certain species in a process known as *eutrophication*.

  Some untreated WW from some industries contain Chlorine compounds and inorganic chloro-amines when discharged into rivers and sea, could have adverse effects on algae, fish and other aquatic invertebrates. Heavy metals like mercury, cadmium, chromium, arsenic, etc. when contained in WW, their presence in rivers, lakes, sea have both chronic and acute toxic effects on aquatic life. Consequently, it is extremely important for WW that contains these elements to be well-treated.

WW from pharmaceutical and cosmetic industries poses adverse effects on aquatic life especially in high concentration if discharge untreated [25a].

2.5. Wastewater treatment process

The WW treatment method seeks to remove all contaminants present in the fresh WW, known as the Influent, that flows into the treatment plant to the barest minimum level in such a way that the liquid that comes out of the treatment, known as the Effluent, is practicably harmless to the environment and humans. Basically, there are four stages in the treatment of WW, they are the preliminary stage (or treatment), the primary, secondary and the tertiary stage.

- **PRELIMINARY STAGE:** The objective at this stage is simply to protect the operational life of the wastewater treatment plant, reduce maintenance cost of the plant and minimize operational problems and prevent unnecessary contamination. This is achieved by eliminating any materials that can clog or damage the pump of the wastewater treatment plant (WWTP) or alter the treatment efficiency of the plant. This stage includes the following:

a) Screening process: It is the first unit operation which the influent is subjected to in WW treatment, the device that is employed is known as *screens* while the materials that are removed from the influent are referred to as the *screenings*. Screenings in most cases are floatable or suspended solids materials which could be paper, plastic, rubber, rags, vegetable matter, etc [26a]. In general, screens are grouped based on the size of the openings and the mechanism of removal: Coarse, Fine and Micro screens.

- ***Coarse screens*** *(Fig. 2)* are distinguished by their large openings; 6-150mm and are usually parallel bars, rods or wires, wire mesh or perforated plates with openings size that could be rectangular or circular in shape. Sometimes it is also known as *bar rack*. Coarse screens could be classified as hand cleaned or mechanically screened screens according to the screening of the wastewater treatment.

Figure 2. *Coarse screen* [27a]

Figure 3. *Static wedge wire screen* [28a]

- ***Micro screen*** *(Fig. 4)* is a filtration device that employs a low-speed rotating drum that has a pre-fixed screening cloth, it backwashes in a gravity flow condition and the openings are in the range of 10-35µm. It is used to remove solids as well as the BOD/COD from the secondary stage effluent, usually before the disinfection of the WW or before the discharge to the environment.

Figure 4. *Micro screen* [29a]

b) Comminutors: *(Fig.5)* They are shredding and screening devices use to capture and reduce the size of solids so as to facilitate the treatment of WW in the downstream process. They can grind solids up to 6-19mm [30].

Figure 5. *Comminutors of wastewater* [31a]

c) Grit removals: Grits are sand, gravel, cinder and other heavy solid materials like eggshells, bone chips, seeds coffee grounds, etc that are denser than the bio-degradable materials contained in the WW. Their presence in WW throughout the treatment operations could cause abrasion in pumps, accumulation in sedimentation tanks and in sludge digesters which could lead to operational difficulties and even blockage in pumps. They are usually removed in grit chambers, examples are Aerated Grit chamber, Detrius tank, Horizontal flow, Hydrocyclone, Votex-type Grit chamber [32a,33a].

d) Fats, oil and grease removal: FOG is a by-product of cooking foods like vegetable oils, dairy products, meat, etc. FOG and other greasy materials like non vegetable oils, wax, soaps, etc. get into WW, most often through domestic and industrial sources. They are usually removed in the skimming tank *(Fig. 6)*. The skimming tank is a chamber designed in such a way that FOG and other similar materials rise and float to the surface of WW until their removal. Compressed air is blown from the diffusers which are usually placed at the bottom of the tank, so that oily materials are pushed upwards. The compressed air serves as coagulants, coagulating greases, soap, etc to form a product known as SCUM on the surface. The scum is then skimmed off the tank.

Sometimes chlorines gas is blown in along the air as it destroys the colloidal effects of protein increasing the removal of oil and other greasy materials by as much as 3000 times [33a,34a].

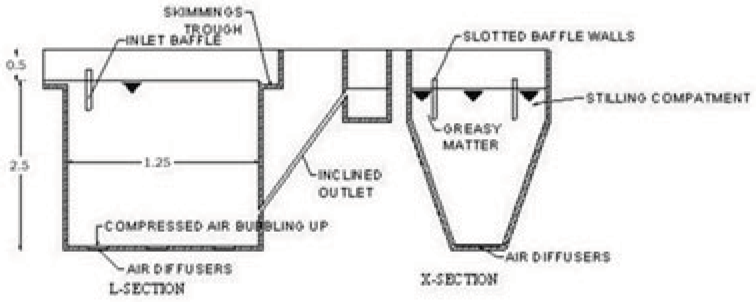


Figure 6. *Skimming tank for wastewater treatment* [35a]

Oil and other greasy materials because they considerably reduce the efficiency of the sludge treatment process, they reduce the biological activity of bacteria and other protozoans, they interfere with the operation of the trickling filter and consequently inhibiting the biological growth of bacteria, etc.

- **PRIMARY STAGE TREATMENT**: The objective at this stage of wastewater treatment is the removal of settleable inorganic and organic solids. The influent from the skimming tank flows into a settling tank known as the **primary clarifier** at a very slow velocity. The slow velocity enables the finer particles present in the influent either settles down the tank or floats upwards to the surface. To achieve this objective, the effluent is left in the tank for 1,5-2 hours, a time that is technical known as the **hydrological retention time** (HRT). The HRT depends on the nature of putrescibles solids to be separated, among other factors. The settleable solids at the bottom begins to accumulate forming what is referred to as the Primary Sludge, it is usually pumped into the **sludge digester** where activated sludge treatment takes place.

  The primary clarifier tank could be circular *(Fig. 7)* or rectangular and to aid the rate of sedimentation of the settleable solids coagulants and flocculants are usually added to the effluent, examples are aluminium sulphate (alum), Iron (III) chloride, etc. Approximately 25-50% BOD, 50-70% TSS and 65% FOG are removed at this stage [37a,38a].

Figure 7. *Circular primary clarifier tank of a wastewater treatment plant* [36a]

- **SECONDARY STAGE TREATMENT:** This is the stage in which the objective is to remove bio-degradable dissolved and colloidal organic solids, as well as the suspended solids that are still present in the effluent from the primary stage. These solids are removed through aerobic biological process that is made possible with the presence of aerobic micro-organisms, principally bacteria. The removal is carried put through any of the following biological processes:

  a) Bio-filtration: This method makes use of sand filters, contact filters and trickling filters for the removal of degradable solids as well as the suspended ones. Although one might tend to think the removal is done by filtration due to the name of the process, it's actually carried out through bio-degradation by the micro-organisms attached to the filter. The trickling filter is most used in wastewater treatment because it's the most effective with stones and plastics used as the coarse media.

  b) Activated sludge process: It is the method that is mostly employed in secondary treatment. The effluent from the primary stage flow into an aeration tanks which serves as the bio-reactors. The effluent is mixed vigorously with the aeration device which supplies oxygen creating a good atmosphere for the bacteria to feed on the organic matter of the effluent and by so doing form flocs that float on the surface of the wastewater. The constant supply of oxygen promotes the multiplication of bacteria through cell division creating activated sludge that floats downwards to the bottom of the basin. It is this process that is referred to as the activated sludge process. At this stage, the effluent looks like a "boiling hot chocolate" *(Fig. 8)* due to the colour of the effluent and the constant supply of air at the bottom.

Figure 8. *Aeration tank of the secondary stage treatment* [39a]

The HRT in the tank is about 8 hours thereafter the liquid flows into the *secondary clarifier (Fig. 9)*. The secondary clarifier is nothing more than a settling tank and it's operated just like the primary clarifier. A part of the activated sludge that is at the bottom of the clarifier is pumped to the sludge digester this part is referred to as *the secondary sludge*. The other part is returned into the aeration tank. At this stage, 80-90% BOD and about 85% TSS is expected to have been removed [39a].

Figure 9. *Secondary clarifier of the secondary stage treatment* [39a]

- **TERTIARY STAGE TREATMENT**: The aim of this stage in WW treatment is to remove all the pollutants contained in the WW to a level that complies with the domestic, industrial and environmental discharge standards into surface water bodies. It involves processes that seek to remove residual suspended solids, toxins, nitrogen and phosphorous containing inorganic compounds, heavy metals containing inorganic compounds like lead, Pb^{2+}, cadmium, Cd^{2+}, and most importantly, pathogens like bacteria, viruses, and parasites like *Giardia*, *Cryptosporidium*, etc. [41a]. It is sometimes referred to as *effluent polishing*.

  The stage begins with filtration, which can be done either by micro screener or sand filtration to remove the suspended solids that could still be present in the effluent from the secondary stage treatment. Activated carbon is usually applied when the objective is to remove toxins. The next objective is the removal of heavy metals. This is done by the precipitation of their compounds, by adding hydrogen sulfide, H_2S:

$$Cd^{2+}_{(aq)} + H_2S_{(g)} \rightarrow CdS_{(s)} + 2H^{+}_{(aq)} \quad \textbf{(2)}$$

$$Pb^{2+}{}_{(aq)} + H_2S_{(g)} \rightarrow PbS_{(s)} + 2H^{+}{}_{(aq)} \quad \textbf{(3)}$$

The phosphate inorganic compounds on the other hand is precipitated by the addition of alum or alternatively the addition of Ferric salts:

$$Al^{3+}_{(aq)} + PO^{3-}_{4\,(aq)} \rightarrow AlPO_{4(S)} \quad \textbf{(4)}$$

The nitrates that are still present are mostly inorganic since it is expected that all the organic nitrates would have been decomposed in the biological process of the secondary stage treatment. Nitrates are highly soluble in aqueous solution and so it’s precipitation of its salts is definitely not the best means of its removal. Another method that can be used is the Ion exchange method:

$$X - OH_{(aq)} + NO^{-}_{3(aq)} \rightarrow X - NO^{-}_{3(aq)} + OH^{-} \quad \textbf{(5)}$$

The final operation in this stage is disinfection. Disinfection can be done through three methods: chlorination, ozone and ultra-violet method. Chlorination is the most common method in use. The disinfection process aims to kill all micro-organisms as well as parasites before it is discharge to surface water bodies. A chlorine solution is injected into the wastewater at the head end of a chlorine contact basin, the amount injected depends on so many factors like the quantity of wastewater, the strength, the source, the pH, the organic content, etc. HRT is usually between 30 minutes to 2 hours and dosage of 5-15 mg/l. Leaving chlorine in wastewater after chlorination is harmful to aquatic life and could even reduce the quality when in high dosage, so the wastewater has to pass through dichlorination. To achieve this, sodium bisulfite is added to the water. Finally, the parameter of the water is checked, the BOD, TSS, and very importantly, its chlorine content, then the water is discharge to the environment or reuse for other purposes [42a,43a].

Fig. 10 shows an overview of the wastewater treatment process.

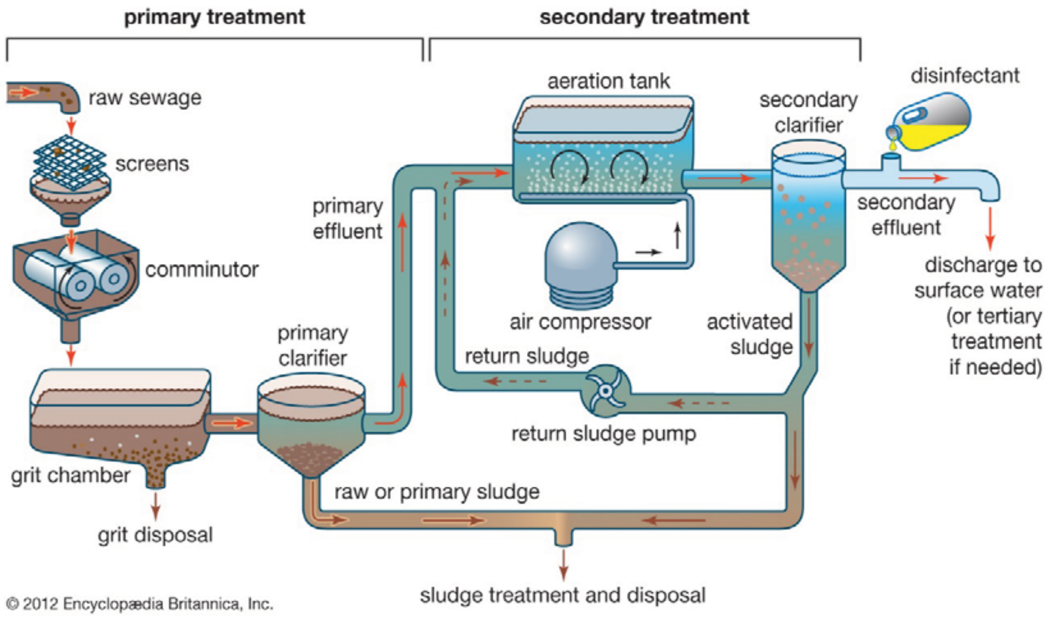


Figure 10. *An overview of the entire treatment process of wastewater* [40a]

3. CONSTRUCTED WETLANDS

As noted in the previous chapter, the harmful impact of discharging untreated WW or even partially treated ones on aquatic life and its direct and indirect consequence on human has led many governmental agencies to enact laws regulating wastewater discharge into the environment. The more new discoveries are made on the negative consequence of discharge of wastewater especially those ones that affect climate change, the more stringent environment laws on discharge become. The consequence of all this is that final effluent of wastewater treatment plant must be of higher quality, the higher the degree of water quality the higher the advancement of the treatment process something that makes the energy, manpower, operational and maintenance costs even higher.

The conventional method of treating WW has always been complex, sturdy, and expensive so much so that a lot of research is being carried out to expand the advantages of non-conventional method over its disadvantage. Some non-conventional method as already mentioned in chapter 1 includes waste stabilization ponds, up-flow anaerobic sludge blanket (UASB), oxidation ditches and constructed wetlands (CW). All these non-conventional methods have their advantages and disadvantages, but CW have been more interesting to research investigators especially in the developing countries because of the following:

a) It uses natural biological processes.
b) It is relatively simple in operation and maintenance when compared to the conventional methods.
c) It does not require additional energy from fossil fuels, something which makes it eco-friendly.
d) It’s not expensive and is relatively simple to design because of its low mechanical technology when compared to the design of the treatment plants of conventional method.

Wetlands are ecotones between terrestrial and aquatic system with a unique hydrologic condition. It’s a term used to describe any wet environment like swamps, marshes, floodplains, peatlands, slough, bogs, fens, muskegs, potholes, and mires. In the past couple of decades, it has been found by research investigators that the function of wetlands goes beyond its use as discharge site of waste; as it was the practice in ancient times, it was observed that it can transform and store nutrients and organic matter through natural biological processes [5]. It is this function of wetlands that led to the creation of Constructed Wetlands, CW of course after decade of research by leading scientists like Dr Keith [6].

Constructed wetlands, CW, are planned engineered system that are designed to treat wastewater using natural physical, chemical and biological processes of a natural wetland in a controlled medium involving many components like soil, sand/gravel, micro-organisms and macrophytes. They are also known as artificial or manmade wetlands. CW are used as an alternative of the biological and chemical stage of WW treatment and are usually designed to stimulate natural processes that occur in natural wetlands but carried-out in a controlled environment.

CW are used to remove suspended solids, pathogens, organic matter, faecal bacteria, nutrients, heavy metals, surfactants, pharmaceutical and personal care products, PPCP, petroleum refinery waste, compost and landfills leachates, fish-pond waste, industrial waste that have already passed through the preliminary and primary treatment like seafood discharge, paper and pulp waste, etc. . They are used solely to treat some wastewater but sometimes are part of a long line of treatment processes).

3.1. Types of constructed treatment wetlands

CW can be classified using different parameters:

- Based on the hydrology: Surface flow CW (SCW), and Subsurface flow CW (SSCW). *(Fig.11)*
- Based on the types of the growing form of macrophytes plants: Emergent plants, Submerged plants, Free floating plants and Floating-leaved plants. *(Fig. 12)*
- Based on the flow path: Horizontal Subsurface flow constructed wetland (HF-SSCW) and Vertical Sub-surface flow constructed wetland (VF-SSCW). *(Fig.11 and 13)*

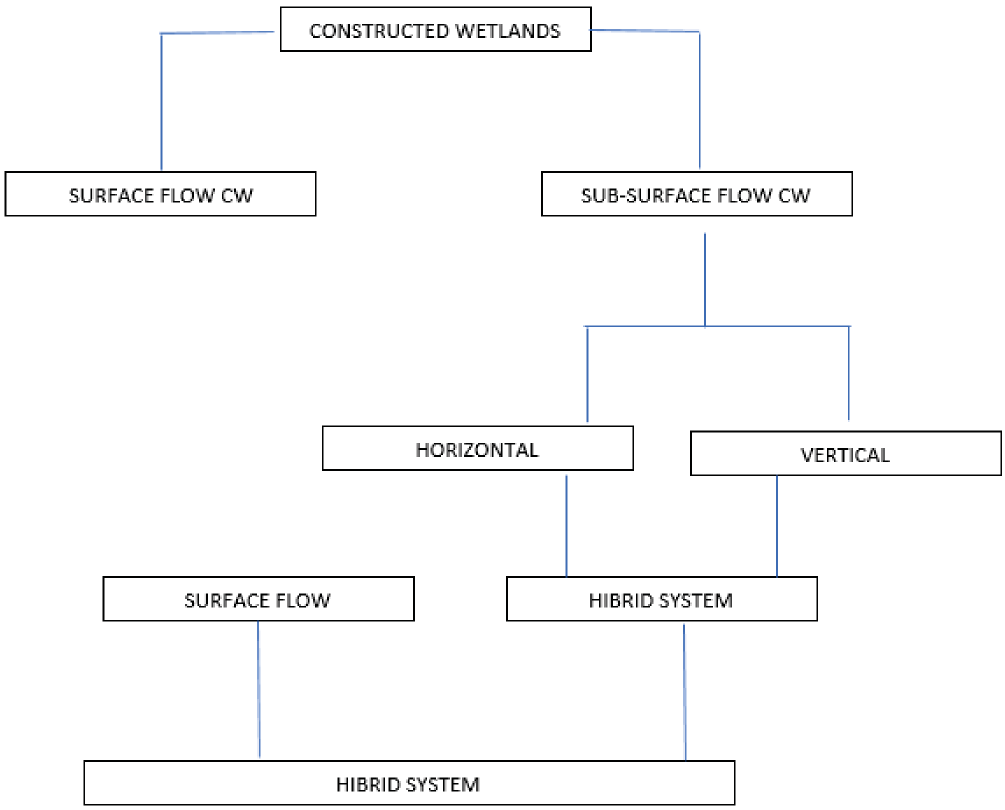


Figure 11. *Types of constructed wetlands*

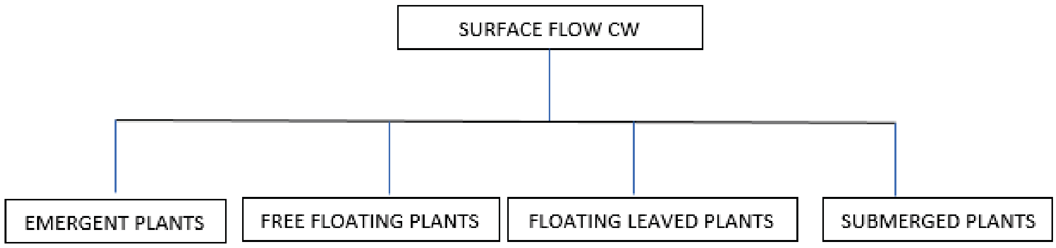


Figure 12. *Types of constructed wetlands according to the macrophytes planted*

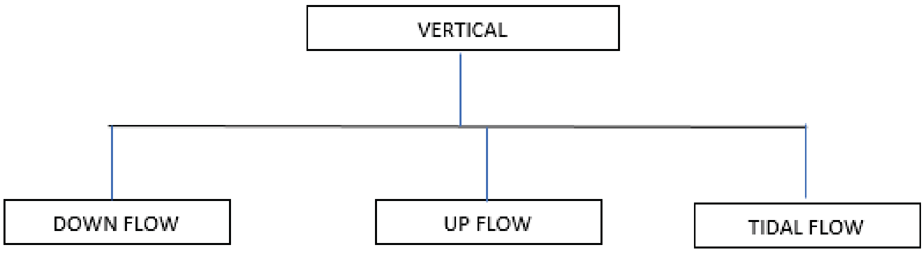


Figure 13. *Types of vertical flow sub-surface constructed wetlands*

To optimize the treatment of WW, most times various types of constructed wetlands are combined to form an **hybrid system**. The hybrid system combines the specific advantage which each type of CW offers, optimizing the treatment process. The hybrid system is usually formed combining HF-SSCW + VF-SSCW; but there are other hybrid systems of this combination, like SCW+ HF-SSCW.

3.2. Components of a constructed wetland

The components of a CW can be grouped into three:

- Wetland macrophyte system
- Wetland media
- Wetland organisms

A. WETLAND MACROPHYTE SYSTEM: Macrophytes refers to all larger aquatic plants growing in wetlands which includes aquatic vascular plants (ferns and angiosperms), larger algae with visible tissue and aquatic mosses, angiosperms dominate the macrophyte system. The presence of macrophytes in wetlands, whether natural or constructed, is often used to classify the wetlands and in fact could determines how effective the wastewater treatment process would be. By nature, macrophytes are photoautotrophic; they make use of solar energy to produce organic compounds which serves as nutrients for micro-organisms present in the wetland [7].

Macrophytes in wetlands can be classified into four groups:

1. **Emergent macrophytes:** These are macrophytes that dominates all other types in wetlands, they grow in shallow waters, banks of ponds, lakes and in shallow marshes. Unlike submerged macrophytes, they are rigid and does not need any support while growing. They can grow between 50-150cm of water depth , adapt very well from a morphological point of view in water-logged or substrate saturated soil due to the large internal air paces which they have for the transportation of oxygen to the root and lastly to the surrounding rhizosphere, this facilitates the degradation of contaminants contained in wastewater. Examples include *Phragmites australis* (Common reed), *Glyceria spp.* (Mannagrasses), *Typha spp.* (Cattails), *Zizania aquatica* (Wild Rice), etc. *(Fig. 14)*

Figure 14. *Typha latifola, an emergent macrophytes* [45a]

2. **Floating leaved macrophytes:** They are also photo-autotrophic with root that can extend to 0,5-3,0m, examples include *Nymphaea spp.*, *Nuphar lutea*, *Nymphoides peltate*, *Potamogeton*, *Polygonum hydropiper*, etc. *(Fig. 15)*

Figure 15. *Flating leaved macrophytes* [46a]

3. **Submerged aquatic macrophytes:** Just like the previous macrophytes they are autotrophic, but they have their photosynthetic tissue under the water while the flowers are on top of the water level. Examples are *Myriophyllum spicatum*, *Ceratophyllum demersum*, *Rhodophyceae*, etc. *(Fig. 16)*

Figure 16. *Submerged macrophytes* [47a]

4. **Freely floating macrophytes:** As the name indicates, they float freely on surface water. Through the process of denitrification, freely floating macrophytes eliminate nitrogen and phosphorus, examples include *Lemna minor*, *Eichhornia crassipes*, *Pistia spp.* like water lettuce, etc. *(Fig. 17)*

Figure 17. *Free floating macrophytes* [48a]

Most soils are fertile ground for the growth of micro-organisms, habitat of some animals like rats, insects, ants, earthworms, etc. but also one for the growth of plants. Soils are generally well drained with air filled pores connecting to the root and rhizomes of plants in such a way that the abundance oxygen-rich air in the atmosphere replenish the air in the soil through rapid diffusion and convection.

In wetland soil, the opposite is the case as the soil are partially or completely water-logged. The pore spaces connecting the soil with the root and rhizome of plants are filled with water creating an anoxic or anaerobic zone for plants as well as animals, micro-organisms. Through research, it has been found that the rate of diffusion of oxygen in air is 106 times faster than in water, something that has been attributed to low solubility of oxygen in water as well as its low diffusion coefficient in water[7]. Consequently, only plants with internal spaces in its organs like leaves, stem, etc for the diffusion of oxygen from the atmosphere into its roots and rhizomes can adapt in this condition.

Aquatic macrophytes have internal air spaces that allow diffusion of oxygen to its roots and rhizomes making it morphological adapted to survive in water- logged condition *(Fig. 18)*.

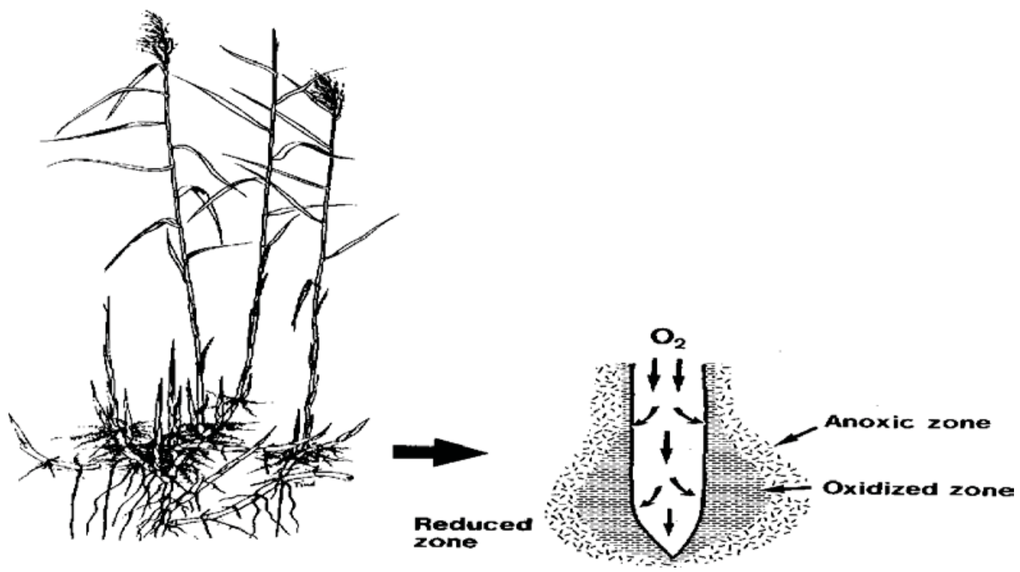


Figure 18. *Sketch of the common reed, phragmites austalis. The oxygen leakage from roots creates oxidized conditions in the otherwise anoxic substrate and stimulates both aerobiv decomposition of organic matter and growth of nitrifying bacteria* [7]

B. WETLAND MEDIA: They are gravel, soil, sand and some organic solid compounds like charcoal, etc. They serve as support to the wetland soil, functions as sites for bio-chemical, physical reactions during the complex wastewater treatment process.

C. WETLAND ORGANISMS: Just like humans, wetland micro-organisms need nutrients to grow and replicate, water, to say hydrated and prevent loss of cellular components and of course a cosy environment that will guarantee the supply of these nutrients, maintained, or at least prevent the variation of the physical condition like pH, temperature, dissolved oxygen, etc. from getting out of range. Wetlands provide this nearly ideal condition which explains the presence of different micro-organisms like Bacteria, fungi, algae, protozoans, etc. [8]

Bacteria are unicellular prokaryotic organisms which decompose both organic and inorganic contaminants in constructed wetlands. They are most often classified according to their mode of nutrition. Bacteria that are capable of synthesizing cell components in the presence of sunlight, referred to as Phototroph, or through chemical reaction of organic compounds present in the environment, in this case referred to as Chemotroph, those that are capable of synthesizing cell components using CO_2 as their major or sole carbon-source are collectively grouped as **autotrophic bacteria**. As one would imagine not all bacteria are autotrophic, some are **heterotrophic**; they are bacteria that source their carbon-source from organic compounds.

1. **Autotrophic bacteria**: They synthesize their nutrients from inorganic compounds like CO_2, H_2S, H_2O, salts, etc. They are sub-divided into other groups:

   i. **Photo-autotroph**: They are capable of using energy from the sun to transform CO_2 and H_2O into organic polymers like carbohydrate, proteins, etc in a process known as photosynthesis and with oxygen as a by-product. These bacteria have chlorophyl pigment in their cell and some of them are anaerobes, examples are cyanobacteria. Photo-autotroph could be purple sulphur bacteria;

they use sulphur compound in the presence of sunlight without water, examples are *Chromatiium, Theopedia, Thiospirilium*, etc., green sulphur bacteria; these ones use hydrogen sulphide in the presence of sunlight, examples are *Chlorobium limicola, Chlorobacterium,* etc, others are non-purple suphur bacteria, non-green sulphur bacteria.

ii. **Chemo-autotroph:** They lack pigment in their cell and so make their nutrients through chemical reactions with inorganic compounds in the presence of oxygen. They also have sub-groups like *Sulphomonas* (sulphur bacteria), *Hydromonas* (hydrogen bacteria), *Ferromonas* (Iron bacteria), *Methanomonas* (Methane bacteria), *Nitrosomonas* (nitrifying bacteria), *Nitrobacter, Carbon bacteria*, etc.

2. **Heterotrophic bacteria**: Most heterotrophic bacteria feed on humans, animals, plants. They are also sub-divided into the following groups:

i. **Photo-heterotroph:** They utilize bio-organic compounds as the carbon source in the presence of sunlight as energy as they have bacteriochlorophyll pigment examples are purple non sulphur bacteria like *Rhodospirilum, Rhodomicrobium, Rhodopseudomonas palustris, etc.*

ii. **Chemo-heterotroph:** They feed on organic compound like protein, carbohydrate, lipids, etc, they could be parasitic bacteria, saprophytic or symbiotic bacteria [49a].

3.3. Components of a constructed wetland

As shown in Fig. 11-13, there are so many types of constructed wetlands, but three threes are mostly used: Free Surface constructed wetland (FSCW), Horizontal flow sub-surface constructed wetland (HF-SSCW), and Vertical flow sub-surface constructed wetland (VF-SSCW).

➢ **FREE SURFACE CONSTRUCTED WETLAND (FSCW):** It's a type of constructed wetland distinguished by the flow of the wastewater to be treated as it is exposed to the atmosphere. FSCW consists of a channel or basin usually lined by an impermeable barrier to prevent the liquid from seeping to surface or underground water bodies. The impermeable layer could be clay or geo-textile [50a] that would be covered with rocks, gravel and soil, with emergent plant like cattais, reeds, rushes, etc growing. *(Fig. 19)*

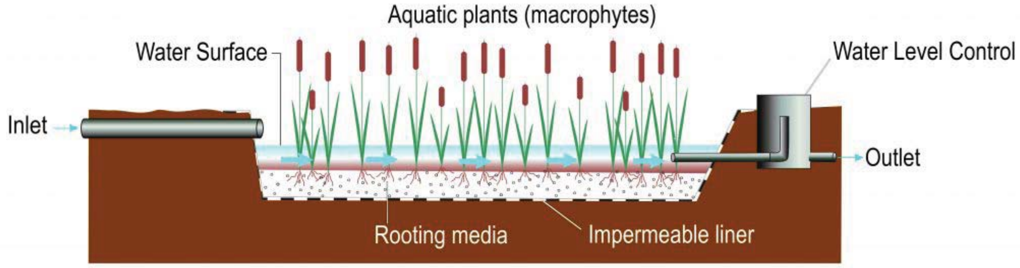


Figure 19. *Free water surface constructed wetland* [44a]

The mechanism of contaminants-removal starts as the wastewater trickles down the wetland, the suspended particles are removed by filtration and sedimentation of heavier particles, the nutrients contained on the particles are also removed as well. Emergent plants and micro-organisms present in the wetland remove nitrogen and phosphorus containing bio-compounds as well as other bio-organic compounds in the wastewater, with precipitates also being formed through chemical reactions, lastly the pathogens are eliminated by decay, by predation of higher organisms and by exposure to direct ultraviolet ray from the sun. All these processes take place simultaneously *(Table 4).*

FSCWs are mostly used as an advanced treatment for effluents from secondary or even tertiary stage treatment.

Table 4. *Advantages and disadvantages of FSWS [50a]*

Advantages	Disadvantages
Aesthetically pleasing and provides animal habitat	May facilitate mosquito breeding
High reduction of BOD and solids; moderate pathogen removal	Requires large area
Can be built and repaired with locally available materials	Long start-up time to work at full capacity
No electrical energy is required	Requires supervision
Problem associated with foul smelling could be solved with good designs and constant supervision	Not appropriate for cold climates
Aesthetically pleasing and provides animal habitat	
High reduction of BOD and solids; moderate pathogen removal	
Can be built and repaired with locally available materials	

- **HORIZONTAL CONSTRUCTED WETLAND (HF-SSCW):** It's a system that consists of a channel or basin with a bed of gravels and sand of a specific size range, a soil from which the emergent plant grows, and the basin is lined with an impermeable layer of clay or geo-textile to prevent leaching. The bed of gravels serves as a filtering medium and also as a growing medium for the micro-organisms in the wetland. The emergent plant usually used in HF-SSCW is reed (*Phragmites spp*), because of its horizontal rhizomes which grow penetrating large area of the system. The macrophytes uses the roots and rhizomes as a substrate for the growth of bacteria and other micro-organisms that are attached to the macrophytes, radial oxygen loss, nutrient intake and as an insulation material of the basin when temperature drops below operating condition. *(Fig. 20)*

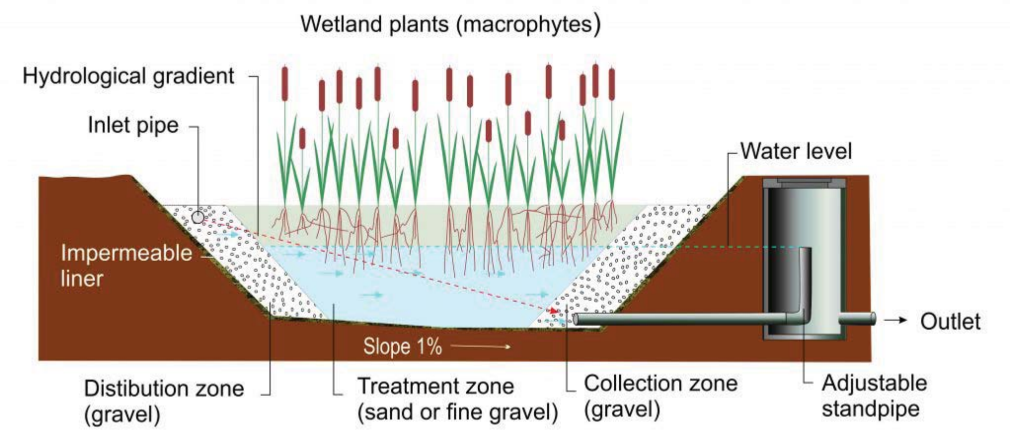


Figure 20. *Horizontal subsurface flow constructed wetland* [44a]

The flow of the influent from previous treatment stage passes through the bed of gravel and sands is maintained below the level of the inert material, creating a ***predominant anoxic environment***, the zone that is close to the root and rhizomes of the macrophytes creates a different environment that is **aerobic** and elsewhere in the bed, a zone that is free from oxygen exuded by the root and rhizomes of the macrophytes and oxygen containing compounds, an **anaerobic** environment emerges. This condition creates a ***redox environment*** of a network of anoxic, anaerobic, and aerobic environment, which is dynamic and versatile, one that allows the growth of several family of specific specie of micro-organism but also one that is harmful for pathogens because of the varying low content of dissolved oxygen.

The treatment of the influent begins as it passes through the filtering media, eliminating suspended particles it contains, the bacteria attached to the media degrades the organic and nitrogen material while phosphorus compounds are removed by adsorption. While the influent flows through the vegetation, microbial degradation continues as well as the removal of phosphorus but this time around through ion exchange method. The removal of phosphorus in HF-SSCW is usually low unless special inert materials are employed [6]. HF-SSCW is used to treat domestic, industrial, agricultural, landfill leachate wastewaters, etc.

- **VERTICAL CONSTRUCTED WETLANDS:** Just like horizontal, it contains a channel or basin, a bed of coarse particles of gravels and sand and a wetland vegetation. The gravels and sand are finer than those of the horizontal because of the need to permit a slow percolation of the influent with the sole objective of having a homogenous distribution throughout the vegetation. A pipe is passed through the filtering media and the soil through which air is passed into the environment. The gravels and sand serve as an attachment for the growth of micro-organisms, a base for the wetland vegetation and more importantly for the removal of suspended particles. The vegetation in VF-SSCW are usually reed (*Phragmites spp*), Cattais (*Typha spp*) and *Echinochoa pyramidalis*. By intermittently dosing the influent, the filtering media pass from an aerobic phase to an anaerobic phase. During a flush, the influent drains downward, the suspended particles are removed by filtration and sedimentation, microbial activity enhanced by aeration degrade the organic and nitrogen particles. The environment is predominantly aerobic in VF-SSCW and consequently rapid nitrification takes place. The major difference between the HF-SSCW and VF-SSCW is not just the flow of influent into the system, it is rather the predominant environment in the two system, in VF-SSCW it is *aerobic* while it is *anaerobic* in HF-SSCW. *(Fig. 21)*

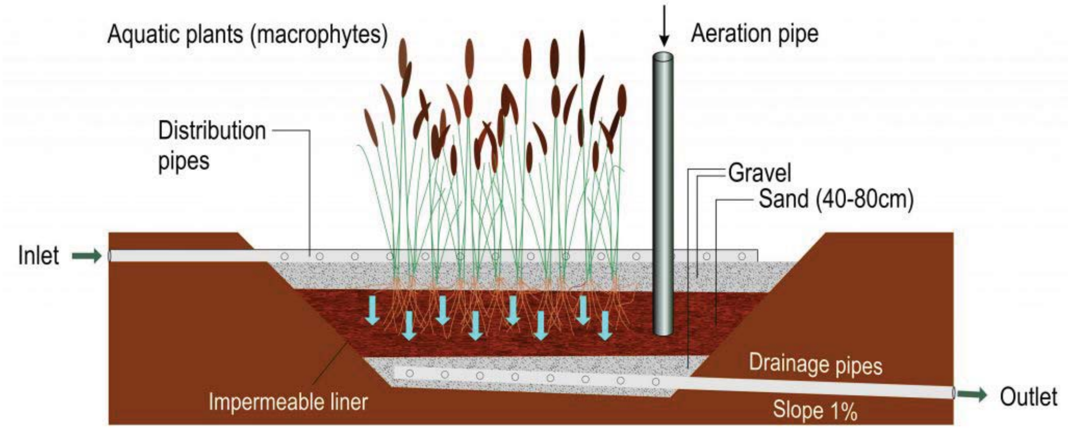


Figure 21. *Vertical subsurface flow constructed wetland* [44a]

4. NITROGEN REMOVAL MECHANISMS IN CONSTRUCTED WETLANDS

Nitrogen is present in wastewater both in organic form and in inorganic form, in organic form it is present as protein; a large bio-polymer of amino acid, as urea; one of the bio-products of the digestion of protein by mammals, uric acid; a bio-product of the digestion of protein by insects, birds, reptiles, etc, chlorophyll; a component of plant cell, etc. The inorganic forms are as many as the varying oxidation state of elemental nitrogen; ammonium; NH_4^+, nitrite; NO_2^- and nitrate NO_3^-, with molecular nitrogen, N_2, nitrous oxide, N_2O, nitric oxide (NO_2 and N_2O_4) and ammonia (NH_3) as the gaseous form.

The removal mechanism of nitrogen in subsurface constructed wetlands is complex as the bio-polymer organic form of nitrogen; the form, which is subjected to microbial degradation, has to be transformed through many forms; forms which are enumerated in the previous section, until it is converted into a simple inorganic compound *(Fig. 22)*. The transformation occurs through many physico-biological processes that starts with **ammonification**.

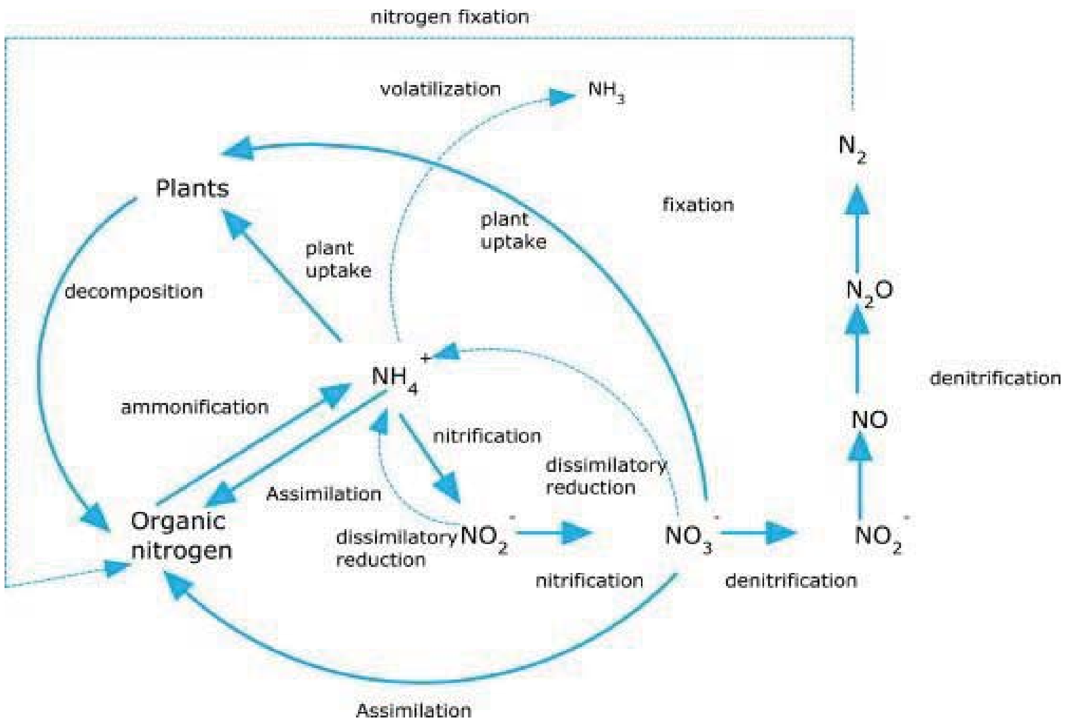


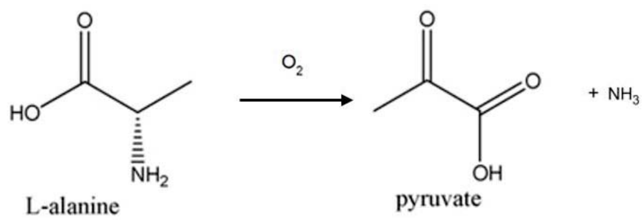
Figure 22. *A schematic diagram of the nitrogen conversion route in sub-surface constructed wetlands* [8]

The removal of nitrogen in sub-surface constructed wetlands is carried out through two ways, the traditional and the Innovative way.

4.1. Traditional nitrogen removal routes

1. **AMMONIFICATION:** Ammonification is the first step of the complex physico-biological process of the removal of nitrogen compounds in constructed wetlands. Nitrogen compounds contained in dead plants, animals, in wastewater are reduced into a simpler nitrogen compounds through the process of AMMONIFICATION. It is a catabolic process where organic nitrogen compound is transformed into nitrogen ammonia; $\mathrm{N\text{-}NH_3}$, or nitrogen-ammonia salt, $\mathrm{N\text{-}NH_4^+}$, in the presence of micro-organisms like bacteria, precisely ammonifying bacteria as these bacteria are known, fungi, and other micro-organisms. The ammonifying bacteria are *Bacillus*, *Clostridium*, *Proteus*, *Pseudomonas*, and *Streptomyces*.

   It's a multi-step exothermic process that include different types of deamination reaction. When ammonification takes place in an oxygen-rich medium, oxidative deamination reaction takes place:



(6)

   Oxidative deamination proceeds:

$$Amino\ acids\ \Rightarrow\ Imino\ acids\ \Rightarrow\ Keto\ acids\ \Rightarrow\ NH_3 \tag{7}$$

   In the soil layer where reductive condition prevails, reductive deamination reaction is what takes, it proceeds in this way:

$$Amino\ acids\ \Rightarrow\ Saturated\ acids\ \Rightarrow\ NH_3 \tag{8}$$

   Ammonification has been found to be faster than nitrification in the zone where oxygen is well enriched, but as the environment changes to anaerobic condition, the rate begins to decrease. The contribution of aerobic process has been found to be small to facultative anaerobic microbial activity. Ammonification is influences by temperature, C/N ratio, types of soil, and nutrients available in the soil, the optimum temperature has been found to be 40-60°C with 6,5-8,5 as the optimum pH [9,10].

2. **NITRIFICATION:** It is the next stage after ammonification provided the concentration of ammonia-nitrogen does not overweigh that of organic nitrogen. It is a bio-chemical process whereby reduced ammonia-nitrogen from organic nitrogen is sequentially converted first to nitrite and later to nitrate in the presence of oxygen and micro-organisms, predominantly chemolithotrophic bacteria. It is a process that takes place in two steps:

$$NH_4^+ + \frac{3}{2}\,O_2 \rightarrow NO_2^- + 2H^+ + H_2O \quad (9)$$

$$NO_2^- + \frac{1}{2}O_2 \rightarrow NO_3^- \quad (10)$$

$$NH_4^+ + 2\,O_2 \rightarrow NO_3^- + 2H^+ + H_2O \quad (11)$$

If the biomass that is generated into consideration as a result of the oxidized $N\text{-}NH_4^+$ or $N\text{-}NH_3$ the equation of the first and second steps become complex.

For the first:

$$\begin{aligned} NH_4^+ + 1{,}9\,O_2 + 0{,}069\,CO_2 + 0{,}0172\,HCO_3^- &\rightarrow \\ \rightarrow 0{,}0172\,C_5H_7NO_2 + 0{,}983NO_2^- + 0{,}966\,H_2O + 1{,}97\,H^+ \end{aligned} \quad (12)$$

For the second:

$$\begin{aligned} NO_2^- + 0{,}00875\,NH_4^+ + 0{,}035\,CO_2 + 0{,}00875\,HCO_3^- + 0{,}0456\,O_2 + 0{,}0875H_2O &\rightarrow \\ \rightarrow 0{,}0172\,C_5H_7NO_2 + 0{,}983NO_2^- + 0{,}966\,H_2O + 1{,}97\,H^+ \end{aligned} \quad (13)$$

The first step is carried out by special nitrifying bacteria; the ammonia-oxidizing bacteria; principally by nitrosomonas, but nirosospirras has also been identified to be present [8], while the second step is carried out by the nitrite oxidizing bacteria (falcultative chemolithotrophic bacteria), again principally by Nitrobacter, others include *Nitrospina*, *Nitrococcus mobilus* and *Nitrospira* [11].

Nitrification is an aerobic biological process s that consumes a lot of oxygen, research has shown that 4,2-4,5 mg O_2/mg NH_4^+ is needed for total oxidation of $N\text{-}NH_3$, which explains the fact that dissolved oxygen, DO, in the media as a factor that affects nitrification. Taking a closer look at equations (12) and (13), there is HCO_3^- among the reactants. The chemical specie is obtained from the following equation:

$$CaCO_{3(s)} + H_2O_{(l)} + CO_{2(g)} \rightarrow Ca(HCO_3)_2 \rightarrow Ca^{2+}_{(aq)} + 2\,HCO^-_{3(aq)} \quad (14)$$

This makes it imperative for $CaCO_{3(s)}$ to be present during the process of nitrification to guarantee the alkalinity of the media. It has been found that 7,13 mg is needed. The optimum temperature is between 30-40°C [10]. Apart from chemolithotrophic bacteria, heterotrophic bacteria have now been known to carry-out nitrification of ammonia nitrogen, examples include *Actinomycetes*, *Arthrobacter globiformis*, *Aerobacter aerogenes*, etc.

3. **DENITRIFICATION:** It is a biological process that takes place in sequence, where nitrate and nitrite are reduced into a further reduced form of nitrogen, usually in gaseous state, in the presence of bacteria. In normal condition bacteria makes use of oxygen as the electron acceptor but other

compounds can be used,like nitrate (or in general nitrogen oxides, Sulphur compounds, etc).These bacteria are technically known as facultative chemolithotrophic bacteria, they belong to group of denitrifying bacteria. The media is usually anoxic with a redox potential, Eh between 350-150 mV , with nitrate or nitrite as the electron acceptors in place of oxygen and also, it takes place in the presence of external carbon source as electron donors. The electron donors could be methanol; $\mathrm{CH_3OH}$, ethanol; $\mathrm{C_2H_5OH}$, glucose, lignin, hemicellulose, etc. The mechanism of the reaction takes place in the following way [12]:

$$\mathrm{NO}^-_{3(aq)} + 2e^- + 2\,H^+ \rightarrow \ \mathrm{NO}^-_{2(aq)} + \mathrm{H_2O}_{(l)} \qquad \textbf{(15)}$$

$$\mathrm{NO}^-_{2(aq)} + 2e^- + 2\,H^+ \rightarrow \ \mathrm{NO}_{(aq)} + \mathrm{H_2O}_{(l)} \qquad \textbf{(16)}$$

$$\mathrm{NO}_{(aq)} + 2e^- + 2\,H^+ \rightarrow \ N_2\mathrm{O}_{(g)} + \mathrm{H_2O}_{(l)} \qquad \textbf{(17)}$$

$$N_2\mathrm{O}_{(g)} + 2e^- + 2\,H^+ \rightarrow \ N_{2(g)} + \mathrm{H_2O}_{(l)} \qquad \textbf{(18)}$$

It is misleading to refer to denitrification as an anaerobic process since the facultative chemolithotrophic bacteria which are involved in the process follow aerobic biological route through the mechanism above, it is in fact an *anoxic process*. Denitrification is affected by redox potential, pH, soil type, absence of oxygen, temperature, presence of denitrifiers, organic matter, etc. Optimum pH lies between 6-8,0 , optimum temperature is 60-75°C with molecular nitrogen $\mathrm{N_2}$ as the principal product when the temperature gets closer to the optimum high temperature $\mathrm{N_2O_{(g)}}, \mathrm{NO_{(g)}}$, are produced when the temperature becomes so low as 5°C. 0,003-1,02$\mathrm{gN/m^2d}$ are the mean value of denitrification rate [9,12].

4. **Plant intake and Biomass Assimilation:** The assimilation of nitrogen in constructed wetlands have been demonstrated in various studies to be one of the means through which nitrogen is removed. Assimilation is a biological process through which micro-organisms and macrophytes convert inorganic forms of nitrogen present in wetlands to organic nitrogen form that form part of the building block of cells and tissues. Nitrogen is most often metabolized as ammonia and nitrate than all other forms by macrophytes [9].

As more nitrogen is metabolized from the system, biomass harvesting ensures that the nitrogen is permanently removed from the WW being treated. Many research studies have shown that plant intake increases during early growth, as a result data of plant intake differ for many research studies [13]. Also, it is important that macrophytes species differ in the type of dissolved nitrogen they metabolize and consequently affect the amount of plant intake, other factors include environmental factors, growth rate, concentration of nutrient in the plant tissue.

Plant intake varies for different studies as pointed out earlier on, 0,6-72 $\mathrm{gN/m^2d}$, 22-88 $\mathrm{gN/m^2d}$, 2-64 $\mathrm{gN/m^2d}$, etc.

Apart from plant intake by macrophytes, micro-organisms also take part in biomass assimilation. They utilize the ammonia nitrogen present in wetlands to synthesize vital cellular components, energy and chemical by-products which are released into the environment. Most microbes are bacteria, fungi, protozoan, etc but most research although still very few in number, are focused on bacteria biomass assimilation.

5. **AMMONIA VOLATILIZATION:** Ammonia volatilization is a physical process in which molecular ammonia is lost to the environment of the constructed wetland through mass transfer. Molecular ammonia is readily soluble in water giving rise to ammonic ion in the medium. The two chemical species exist in equilibrium in the solution and their concentration in the solution depends on the pH on the solution and the temperature. The solubility of ammonia takes place according to the following equation:

$$\mathrm{NH_{3(g)}} + \mathrm{H_2O_{(l)}} \rightleftharpoons \mathrm{NH_4OH}_{(aq)} \rightleftharpoons \mathrm{NH}^+_{4(aq)} + \mathrm{OH}^- \qquad \textbf{(19)}$$

A lower value of pH but higher value of temperature leads to higher concentration of ammonium ion [14].

6. **AMMONIA ADSORPTION:** Adsorption is a separation technique in which molecules of a chemical specie known as the adsorbate deposit on the surface of another substance known as the adsorbent. In constructed wetlands, ammonia ion, $\mathrm{NH}^+_{4(aq)}$, is removed from the wetlands system by using materials that have adsorbent preference for ammonia during the flow of WW in the media, examples of materials commonly used are zeolites, biochar, limestone, etc. More elaboration is made in chapter 6 of this study. Like all adsorption process, adsorption of ammonia nitrogen is a reversible process [15], when the concentration of ammonia in the wetlands system is low, the ammonia adsorbed on the materials used is desorbed to maintain the equilibrium and the opposite holds if the ammonia concentration is too high more ammonia would be adsorbed and sometimes the ammonia adsorbed could also be oxidized to nitrate if the dissolved oxygen level is high [12]. Adsorption follows many isotherm theories like Langmuir adsorption isotherm, Freundlich adsorption isotherm, Temkin adsorption isotherm, etc could be used to model the concentration of ammonia adsorbed on the adsorbent.

   Adsorption of ammonia is influenced by nature and type of soil organic matter, types of macrophytes employed in the wetland system, type and nature of the adsorbent employed, etc.

7. **DISSIMILIATORY NITRATE REDUCTION TO AMMONIA (DNRA):** DNRA is one of the many biological processes in which nitrate is reduced in wetlands. Unlike denitrification and **anammox** (this would be explained in section 4.2) where nitrate is converted into a form that leaves the system, DNRA transforms nitrate first to nitrite and later to ammonium ion in the presence of micro-organisms and an electron carbon donor. At hypoxic condition, DNRA could compete with denitrification for nitrate in such a way that the denitrification rate diminishes. It should be emphasized that in DNRA process the reduced nitrogen form, the ammonium ion, is not lost to the to the environment but retained in the wetland system in a biological reactive form, a form that can be used for biomass assimilation by plants and micro-organisms.

DNRA is caried out by strictly facultative heterotrophic bacteria utilizing organic carbon sources as electron donor but some research studies have shown chemolitho-autotrophic bacteria participating in DNRA process but in this case the nitrate is reduced in the presence of sulfur compounds or inorganic compounds. Factors that affect DNRA are the pH of the media, organic matter concentration, temperature, the growth rate of bacteria that take part in the process, the concentration of sulfide, reduced Iron, ratio of electron donor/ acceptor, etc.

4.2. Short cut nitrogen removal routes

As it was explained in section 4.1 the nitrification process is complex oxic process in which the ammonium ion is converted into a sequential chain of reduced nitrogen forms in the presence of chemolithotrophic bacteria and carbon electron donating source. The consequence of this long reaction mechanism is that:

- A high dosage of $\mathrm{CaCO_{3(s)}}$ is needed to maintain the alkalinity of the media.
- An organic carbon source is needed otherwise the reaction will not proceed to complete oxidation of $\mathrm{NH}^+_{4(aq)}$ with the attendant consequence of the production of nitrous oxide, $\mathrm{N_2O_{(g)}}$, a greenhouse gas.
- Nitrification process is aerobic that needs as much as 4,2-4,5mg O_2/ mg $\mathrm{NH}^+_{4(aq)}$.

As a result of the above reasons and indeed much more, researchers have been looking for a biological process that would reduce the long mechanism of the nitrification process into a sort of partial nitrification in which the reduction of the ammonium ion only gets to nitrite, $\mathrm{NO}^-_{2(aq)}$, the sequential part of the nitrification process where nitrite is oxidized to nitrate, $\mathrm{NO}^-_{3(aq)}$ es eliminated. To achieve this a bio-process in which the nitrite oxidation is inhibited that way the nitrite is left for denitrification, the ammonium oxidizing bacteria (AOB), is made to out-compete the nitrite oxidizing bacteria, NOB. This whole process is sometimes referred to as **partial nitrification**|16**]**. The denitrification process could be carried-out in an aerobic or anaerobic condition.

Novel nitrogen removal routes in which partial nitrification is a part followed by different method of denitrification have since been developed, examples are:

5. **ANAMMOX**: Anammox stands for Anaerobic Ammonium oxidation, it’s a biological process where ammonium ion is converted through many sequential reactions to gaseous molecular nitrogen in the presence of a special group of bacteria, anammox bacteria. The process consists of two parts; partial nitrification carried out by ammonium oxidizing bacteria and anammox process carried out by anammox bacteria. Anammox process offers a lot of advantages over conventional nitrogen removal biological processes as:

    - It does not need an external carbon source for the reaction to go to completion unlike conventional biological processes.
    - It reduces process energy demand as aeration is not needed, very low amount of oxygen is needed.

- It has a higher nitrogen removal rate as much as 2,8-5,7 gN/m^2d and the space requirement for the process is small [8].

The pathways in which nitrite gets converted to molecular nitrogen are many as investigators have not really been able to agree on which of the pathways should be discarded or accepted, but two pathways have always stand out. After the oxidation of ammonium ion to nitrite in partial nitrification, the nitrite could be converted through the following pathways in the anammox process:

- The nitrite is first of all reduced to hydroxylamine , NH_2OH, then it reacts with ammonium ion, $NH^+_{4(aq)}$ to form hydrazine, N_2H_4.
- In the alternative pathway, nitrite is first of all reduced to nitric oxide, NO, it then reacts with $NH^+_{4(aq)}$ to form N_2H_4.

Thereafter, hydrazine is then further reduced to molecular nitrogen, N_2. The overall equation of the global reaction is given as:

$$\begin{aligned} &NH_4^+ + 1{,}32NO_2^- + 0{,}066HCO_3^- + 0{,}13H^+ \rightarrow \\ &\rightarrow 1{,}02N_2 + 0{,}26NO_3^- + 0{,}0066CH_2O_{0,5}N_{0,15} + 2{,}03H_2O \end{aligned} \quad \textbf{(20)}$$

The bacteria responsible for the transformation in the anammox process, anammox bacteria belong to the phylum of Planctomycetes with five genera found to take part in anammox process: *Brocadia*, *Kuenenia*, *Anammoxoglobus*, *Jettenia*, and *Scanlindua* [17]. Although the anammox is found in many natural wetlands, however its growth rate is very low, 0,04-0,006/d at 35°C. This is large due to many inhibition parameters like the presence of various compounds in WW like sulfide, phenol, ammonium ion, nitrite, aldehyde, alcohol, etc and also the biomass yield of anammox is also very low, 0,13g VF-SSCW/g NH_4^+. All these factors limit the application of anammox bacteria to continuous WW treatment processes.

In low oxygen and anaerobic condition anammox process have used to achieve NH_4^+ by as much as 27-49% and 2,4 for nitrogen. Factors that affect anammox process include pH, temperature, nitrogen loading, dissolved oxygen, carbon sources, etc. Anammox is usually applied for WW with high ammonium content like landfill leachate, WW from pharmaceutical industries, WW from monosodium glutamate industries, etc.

6. **SHARON:** SHARON is an abbreviation of *Single reactor for High activity Ammonium Removal Over Nitrite*, it's a biological process where there is a partial nitrification of ammonium ion to nitrite like all novel biological processes followed by the denitrification of the nitrite in an anaerobic condition. In this process, the strategy is to make the nitrite oxidizing bacteria inactive during the partial nitrification by taking advantage of the difference in the microbial growth rate of the two groups of bacteria present during the process of nitrification. There are so many factors that affect microbial rate of bacteria in general, among these factors are pH, the residence time for aeration and temperature.

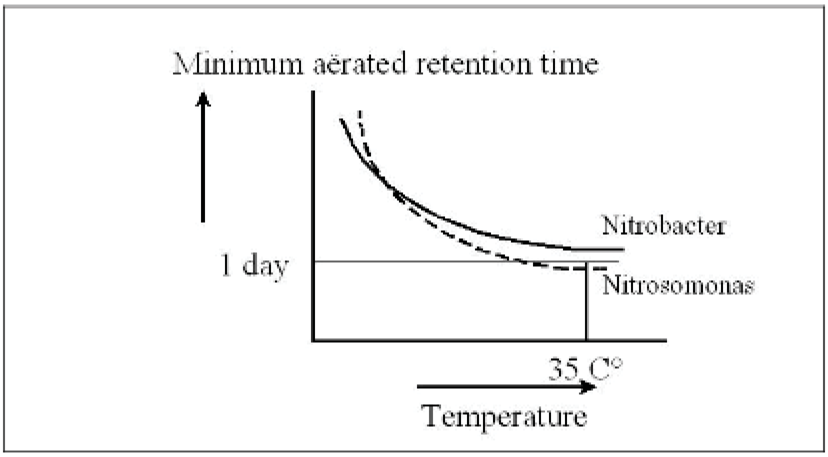


Figure 23. *Different growth rate of ammonia and nitrite oxidizing bacteria* [18]

From fig 23, it can be observed that there is huge difference between the minimum residence time for aeration for both group of bacteria, while that of Nitrosomonas, an ammonia oxidizer is approximately a day while that of nitrite oxidizer is more. Consequently, with the knowledge that the nitrite oxidizing bacteria needs more than a day for to be biological active irrespective of the temperature, the residence time for aeration of the reactor is set for a day while increasing the temperature to close to 35°C and adjusting the pH to optimum value of ammonia oxidizing bacteria.

The end-result is that the nitrite oxidizing bacteria is wash-out of the reactor and the product of nitrification is nitrite, to be followed by denitrification. Anammox is the denitrification process that is usually applied. The overall product is gaseous molecular nitrogen along with other by-products. Sharon is mostly used for high strength ammonia WW like reject water from dewatering of digested sewage sludge, WW from incineration plants, etc.

7. **CANON:** Canon refers to *Completely Autotrophic Nitrogen Removal Over Nitrite*. It is a special kind of anammox process that consists of a combined partial nitrification of ammonium ion followed by the denitrification of nitrite by anammox bacteria in an anaerobic condition and all the biological process taking place in one continuous unit. In this process approximately half of the ammonium ion present in the system is converted to nitrite by ammonia oxidizing bacteria, consuming oxygen in the process that way creating an anoxic condition. The anammox bacteria then utilize the nitrite as an electron acceptor in this anoxic to oxidize the ammonium ion left in the system, converting it to gaseous molecular nitrogen.

The yield of the process would depend on the capability of the two groups of bacteria to exist and complement the sequential reactions. The process needs to control the concentration of dissolved oxygen, enough for the ammonia oxidizers to nitrify while at the same time reducing it to a level sufficient enough to create an anoxic condition for anammox bacteria.

The Canon reaction follows this equation:

$$NH_4^+ + 0{,}85\ O_2 \rightarrow 0{,}435\ N_2 + 0{,}13\ NO_3^- + 1{,}3\ H_2O + 1{,}4\ H^+ \qquad \textbf{(21)}$$

Like SHARON and ANAMMOX, Canon is more environmentally friendly and economical than the conventional biological process to remove nitrogen.

Not all these processes both biological and chemical, are nitrogen removal processes, some are just transformation processes. Processes like Ammonification, nitrification, dissimilatory nitrate reduction, are transformation process where the total nitrogen, TN, in the wetland system remains constant while on the other hand Nitrification, biomass assimilation, adsorption (depending on the aerobic/anoxic/anaerobic condition), plant uptake, volatilization and all the novel nitrogen processes are all removal processes. Nitrification-denitrification still remains the major nitrogen removal process in wetlands system.

5. ORGANICS REMOVAL MECHANISM IN CONSTRUCTED WETLANDS

Although most organic matter in WW contains nitrogen and also phosphorus, there are some that do not contain these two elements. These organic materials are in WW in two forms, as solid known as particulate organic material, POM, or as dissolved matter, better known as Dissolved organic matter, DOM. The removal of organic matter starts with the filtration and sedimentation of POM by the rhizosphere of macrophytes as well as the soil in the wetland, they then start to accumulate, disintegrate, and hydrolyse. The hydrolysis of the retained POM yields simple monomers of glucose, fatty acids, amino acids, xylose, hydrogen, etc. all these monomers dissolving to form part of DOM [19].

DOM are reduced through two pathways, aerobic and anaerobic degradation.

5.1. Aerobic degradation

Aerobic degradation is the breakdown of monomers of organic matter in the presence of oxygen and micro-organisms to produce in most cases carbondioxide and water. Chemoheterotroph are the micro-organisms involved in the degradation where they use oxygen as the electron acceptor. For this process to take place in wetland the environment has to be aerobic, a condition that is fully guarantee in VF-SSCW. An example of a bio-degradation reaction is given below:

$$\mathrm{C_6H_{12}O_6} + 6\ \mathrm{O_2} + microbes \rightarrow 6\ \mathrm{CO_2} + 6\ \mathrm{H_2O} + biomass \qquad \textbf{(22)}$$

Aerobic degradation is highly influenced by the level of dissolved oxygen in wetland, the amount of dissolved *biodegradable* organic matter and the strength of WW being treated. Research has shown that the BOD/COD of WW gives an indication of the level of the proportion of the bio-degradable organic matter present, 0,3 indicates that it contains organic matter that are difficult to be biodegraded, 0,5 that it can be easily bio-degraded [8]. The presence of oxygen and the high redox potential of the environment makes aerobic degradation faster than anaerobic [20].

5.2. Anaerobic degradation

Due to the very solubility of oxygen in aqueous medium, 40 mg/l and the radial oxygen loss in the rhizospheres of macrophytes, different points in sub-surface constructed wetlands are always changing, the surface water level, few centimetres below the surface water level and also, the medium close to the rhizospheres of the macrophytes, all these environments are aerobic. As one moves deep down the wetland, the environment begins to change from aerobic to anoxic and further deep down, especially inside the wetland soil, the environment is completely anaerobic. *(Fig. 24)*

As anaerobic bacteria are present in anaerobic environment which ensure that the process of anaerobic biodegradation takes place. Anaerobic biodegradation is a series of processes *(Table 5)* in which organic matter are broken down by micro-organisms in the absence of oxygen. It is usually carried out by heterotrophic bacteria. The first reaction in anaerobic biodegradation is fermentation and it is then followed by any of the following reactions or a combination of them taking place in series.

Fermentation is a process in which acid forming bacteria convert dissolved organic matter into organic acids, organic alcohol in an anaerobic environment. The process occurs according to the following reactions:

$$\mathrm{C_6H_{12}O_6} + microbes \rightarrow 2\ \mathrm{CH_3CH(OH)COOH} + biomass \tag{23}$$

$$\mathrm{C_6H_{12}O_6} + microbes \rightarrow 2\ \mathrm{C_2H_5OH} + biomass \tag{24}$$

The degradation continues with other different group of bacteria and the next process depends on the ease with which the compound would serve as an electron acceptor would determine the next bio-process. The last process in a anaerobic biodegradation is Methanogenesis which takes place as follow:

$$4\ \mathrm{H_2} + \mathrm{CO_2} + microbes \rightarrow \mathrm{CH_4} + 2\ \mathrm{H_2O} + biomass \tag{25}$$

$$\mathrm{CH_3CO_2^-} + 4\ \mathrm{H_2} + microbes \rightarrow 2\ \mathrm{CH_4} + \mathrm{H_2O} + \mathrm{OH^-} + biomass \tag{26}$$

Sulphate reduction reaction removing organic matter in the presence of bacteria:

$$\begin{aligned} &2\ \mathrm{CH_3CH(OH)CO_2^-} + \mathrm{SO_4^{2-}} + \mathrm{H^+} + microbes \rightarrow \\ &\rightarrow \mathrm{CH_3CO_2^-} + 2\ \mathrm{CO_2} + 2\ \mathrm{H_2O} + \mathrm{HS^-} + biomass \end{aligned} \tag{27}$$

$$\begin{aligned} &\mathrm{CH_3CO_2^-} + \mathrm{SO_4^{2-}} + \mathrm{H^+} + microbes \rightarrow \\ &\rightarrow 2\ \mathrm{CO_2} + 2\ \mathrm{H_2O} + \mathrm{HS^-} + biomass \end{aligned} \tag{28}$$

Nitrate reduction reaction removing organic matter:

$$\begin{aligned} &\mathrm{C_6H_{12}O_6} + \mathrm{NO_3^-} + microbes \rightarrow \\ &\rightarrow 6\ \mathrm{CO_2} + 6\ \mathrm{H_2O} + 2\ \mathrm{N_2} + 4\ \mathrm{e^-} + biomass \end{aligned} \tag{29}$$

Iron reduction reaction removing organic matter:

$$\begin{aligned} &\mathrm{CH_3CO_2^-} + 8\mathrm{Fe^{3+}} + 3\mathrm{H_2O} + microbes \rightarrow \\ &\rightarrow 8\ \mathrm{Fe^{2+}} + \mathrm{CO_2} + \mathrm{HCO_3^-} + biomass \end{aligned} \tag{30}$$

In between hydrolysis and methanogenesis, there are three bio-processes where the intermediate products are further reduced, they are Acidogenesis and Acetogenesis. In acidogenesis, volatile fatty acids, ammonia, hydrogen sulfide, carbon dioxide, ammonia, etc are produced from the product of hydrolysis while in the case of acetogenesis, acetic acid, carbon dioxide and hydrogen are generated.

Table 5: *Microbial groups involved in various redox reactions in wetland soils*

	Redox potential (mV)	Electron acceptor	Decomposition products	Microbial groups
Aerobic	300	O_2	CO_2	Aerobic fungi and bacteria
Fermenting	-100 - 300	Organics	Organic acids, CO_2, H_2, alcohols, aminoacids	Fermenting bacteria
Facultative anaeorobic	100 - 300	NO_3^-, Mn^{4+}, Fe^{3+}	N_2O, N_2, CO_2, H_2O Mn^{2+}, CO_2, H_2O, Fe^{2+}, CO_2, H_2O,	Denitrifying bacteria, Mn^{4+} reducers Fe^{3+} reducers
Obligate anaerobic	less than -100	$SO_4^{2+,}$ CO_2 and acetate Organic salts	HS, CO_2, H_2O, CH_4, CO_2, H_2O, acetate, CO_2, H_2	Sulphate reducers Methanogens H_2 producing bacteria

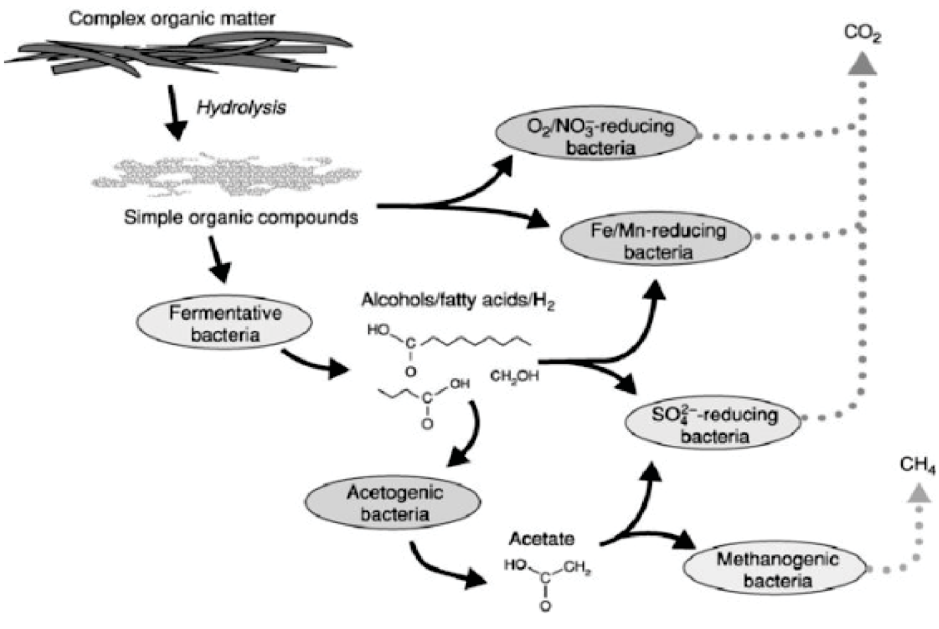


Figure 24. *Pathways of organic decomposition in wetland soils* [21]

HF-SSCW is the type of sub-surface constructed wetlands where predominant anoxic and anaerobic condition are produced consequently, the above processes of anaerobic bio-degradation are expected to take place. Many investigative research-works, [22,23,24] have detected volatile fatty acid as well as methane among the component of effluent of HF-SSCW, confirming that anaerobic biodegradation has taken place. Anaerobic process also takes place in VF-SSCW but in insignificant proportion and anaerobic bio-degradation is hugely dependent on redox potential.

6. INFLUENCE OF ENVIRONMENTAL FACTORS AND OPERATIONAL CONDITIONS ON NITROGEN AND ORGANICS REMOVAL IN SSCW

All the processes that are involved in the removal mechanisms of nitrogen and organic matter takes place in an environment that depends on various parameters of the wetlands. These parameters of the constructed wetlands are themselves depended on the physical, chemical, and biological properties of the incoming WW to be treated. Parameters like pH, temperature, the Dissolved Oxygen level, DO, presence of organic carbon source, and operation parameters like Hydraulic loading and Retention time, mode of influent feed, nitrogen and organic loading, plant harvesting recirculation and of course the nature of wetland media employed.

6.1. Wastewater pH

Denitrification is one of the major removal routes of nitrogen from WW in sub-surface constructed wetlands and a closer look at equations 12-15 shows that hydrogen proton is consumed in the biological process, this means that the pH of the influent would affect the removal of nitrogen during the treatment process. Denitrification process on the other hand is also dependent on nitrification, a close look at equations 6,7-11 shows that $CaCO_{3(s)}$ is consumed producing H_2CO_3(aq), a weak acid. This means that the weak acid condition produced during the nitrification process is neutralized by the consumption of proton in the denitrification process, giving rise to a kind of buffer solution where the pH of the environment is a bit stabilized. To stabilize the pH of the environment, the nitrification and denitrification processes must take place at the same rate something that is difficult to control. Consequently, a faster nitrification rate could result in a substantial drop in the pH of the environment hampering microbial activities. If the opposite scenario takes place where the denitrification out-paces nitrification, there would be a substantial increase in the pH, again negatively affecting microbial activities.

The long-time effect of a sharp drop or increase in pH reduces immensely the efficiency of the nitrogen removal process. Research works have shown that the optimum pH range for denitrification is between 6,0-8,0 but the best yield 6,5-7,5; slightly acidic [25].

The pH of the influent also has an effect on the removal of organic matter during the bio-degradation process. Although no recent research work has shown that the pH of the environment has an effect on the bacteria activity during aerobic degradation of organic matter, it has however been confirmed that pH affects the activity of methano-gen bacteria during the process of methanogenesis in anaerobic biodegradation. The optimum pH range for methano-gen bacteria is 6,5-7,5 [8].

6.2. Temperature

Generally, temperature affects metabolism of micro-organisms and so it isn’t any surprise that it does affect the various physical and bio-chemical processes that occur in sub-surface constructed wetlands, especially the major removal routes like denitrification. Research works have shown that the optimum values obtained during nitrification take place within a temperature of 16-32°C [26,27] while 20-25°C is the optimum range for denitrification [25].

Research works performed at different climatic period confirms the effect of temperature on nitrogen removal, Bilal Tunsciper noted a better removal rate for NNH_4^+ and NNO_3^- during summer than in winter, Langregarber et al found out that there was a significant reduction in the removal efficiency of NNH_4^+ when then operating temperature was below 12°C. Jaime Nivala and her team in their research found that there is a significant different in the removal of NNH_4^+ when the research was carried out in the summer than in the winter. All these point that there is a direct link between the temperature of constructed wetlands and the various microbial activities [28-30]. To guarantee that constructed wetlands keeps operating at an optimum level, researchers sometimes use a mixture of constructed wetlands media like saw dust, washed gravels, sand, pea gravel as an insulating layer [8].

6.3. Dissolved oxygen

In sub-surface constructed wetlands, oxygen is an important factor in the removal efficiency of nitrogen and organic matter especially in vertical constructed wetlands where optimum nitrification takes place. Many of the bio-chemical processes through which nitrogen and organic matter are removed, are reactions which take place by the actions of micro-organisms, some of these micro-organisms need an aerobic condition to carry-out their metabolism, a condition that is met by the presence of dissolved oxygen.

Research has shown that 28 g O_2 /m^2 d is the diffusion rate of oxygen in VF-SSCW, a rate that is not exceedingly sufficient for all the bio-chemical processes taking place, as a result sometimes researchers employ force aeration to bridge the oxygen flow rate deficit [31]. To demonstrate the impact of dissolved oxygen in WW treatment, Ong and his team compared the WW efficiency of two VF-SSCW, one with natural aeration while the other employed a forced aeration. It was found that the VF-SSCW with forced aeration showed 94% for COD and 90% for $N\text{-}NH_4^+$ removal efficiency against 50% and 90% obtained for the natural aeration. This was also confirmed by research works of Ong *et al.* [32] and Stefanakis and Tsihrintzis [33] who obtained high removal efficiency with a better system of aeration of wetland surface.

6.4. Availability of carbon sources

In subsurface constructed wetlands, nitrogen removal is accomplished through plant intake, bio-mass accumulation, ammonia volatilization and denitrification but of all these processes denitrification is the major removal mechanism. Denitrification being a process that takes place in anoxic condition, needs a carbon source that would act as an electron donor, consequently, the efficiency of the process is very often limited by carbon sources which could be organic or inorganic. WW usually contains carbon, mostly organic but they are not always in sufficient quantity for the different bio-chemical processes, as a result external organic carbon are added to the constructed beds or they can be added internally through the use of wetland media rich in carbon *i.e.* internal carbon addition. Researchers have been trying to find the optimum C/N ratio for nitrogen and organic matter removal, Fang et al 2018 used various C/N in their research works to enhance the removal TN of an artificial sewage, they found that at the optimum value C/N= 5 they obtained the highest removal efficiency, 75,4% [34]. Zhao et al also used various C/N ratios , C/N = 2,5, 5 and 10 in the treatment of a simulated sewage using a VF-SSCW, they obtained the highest TN removal for the ratio of 2,5 [35].

For research where the HF-SSCW was used, Rustige and Nolde [36] in the treatment of landfill leachate used a varying amount of C/N between 0,1-0,8 obtained a denitrification rate between 10-75% using acetic acid as the external carbon source. Songliu et al (2009)[37] in the treatment of artificial sewage using HF-SSCW and glucose as the external carbon source, obtained a 20% improvement in the removal of NO_3^-.

From the above scenarios it is obvious that the presence of a carbon source that is readily available during denitrification enhance the removal of nitrogen in various forms, although the amount of its impact varies for different types of constructed wetland and the optimum C/N depends on various factors like the configuration of wetland, the type of macrophytes, the type of nitrogen composition and of course the strength of the WW.

6.5. Hydraulic retention time

Hydraulic retention time (HRT) is the time the influent spends in the constructed wetland, it is usually calculated by dividing the volume of the influent with the volume of the constructed wetland. HRT is an important factor as it determines the period microbes feed on the substrates as well as the period of biodegradation.

A lot of research works have shown that increasing the HRT enhances the removal of nitrogen in wastewater [28] but one has to be careful in not prolonging HRT for too long if the dominating condition in the constructed wetland is anaerobic [8]. The ideal period recommended for HRT is between 2-10 days, that’s because an HRT longer than 2 days does not enhance organic matter removal in HF-SSCW and in general the removal of nitrogen in constructed wetland requires longer HRT than the removal of organic matter [38].

6.6. Hydraulic loading time

Hydraulic loading rate (HLR) is the rate at which WW is delivered into the sub-surface constructed wetland, it is usually expressed in m/day. HLR is one of many factors that affect the efficiency of the treatment process of WW in sub-surface CW. A high HLR means the WW nutrients would pass quickly through all the sub-surface CW components in the media leaving little time for all the bio-chemical processes to take place with the end result of very low treatment especially when the HRT is low. Consequently, researchers have been studying the optimum value of HLR that would not compromise WW treatment efficiency.

For VF-SSCW, Kantawanichkul et al [39] in their treatment of WW from pig farm and domestic sources, observed that increasing values of HL led to reduced value in the nitrogen and organic matter for pig farm WW but in the case of domestic WW, no notable reduced value was noted in their removal. Stefanakis and Tsihrintzis [40] also obtained similar results when they increase the HLR; 0,19, 0,26 and 0,44m/d for the experiment that went on for three years, but in the third year there was an increase in the nitrogen removal 12,1-16,4 $\mathrm{gN/m^2d}$ for N-NH_4^+ and 140-195g COD/m^2 d for organic matter. It could be observed that when the CW employed is VF, the strength of the WW and period in which the CW is operated, time long enough for the macrophytes to have adapted to the changes effected in the variation of HL, would determine if the removal of nitrogen and organic matter would increase. Further research in the future would really be welcomed.

In HF-SSCW on the other hand increase in HL generally reduces drastically the removal of nitrogen and organic matter. This could be due to the fact that in HF little nitrification takes place [8].

6.7. Mode of influent feed

The feed mode of WW into constructed wetlands needs to be well designed in such a way that the influent has the maximum contact with wetlands components. The influent mode refers to the mode in which the fr4esh WW is introduced into the sub-surface CW, it could be continuous, intermittent, batch, tidal or step feed. In the research work of Stefanakis et al [41], various different feed mode was used in the treatment of synthetic domestic WW in a HF wetland and the same WW was treated in a non-step feed using the same sub-surface constructed wetland, they found out that the organic matter removal was more than 87% in the treatment with a the specified feed mode against 82% with the treatment without feed mode.

Also, Shubiao Wu et al [42] were able to prove that using tidal flow for the influent, there is a better improvement of NNH_4^+ and organic matter and even with high hydraulic loading the theoretical oxygen demand was still met during the treatment of an artificial WW. Almost all research works investigating the effect of feed mode on enhancing WW treatment has shown that a better removal efficiency of nitrogen and organic matter as the different feed mode has ensure enhanced aeration during bio-chemical processes taken place in the treatment process.

6.8. Recirculation

Recirculation, when applied to most chemical transformation processes tends to increase the yield, its application to WW treatment in SSCW is not an exception. Various research works have shown that recirculation improves nitrogen and organic matter removal, Manuel Soto et al [43] employed various configurations of a two-step hybrid system to treat simulated food industry WW, the TN removal rate increased up to 73%.

In the work of Sun *et al.* [44], agricultural WW was treated with a VF-SSCW, they obtained 51-77,6% for the removal of organic matter and 19,4% for NNH_4^+ but these data changed when effluent recirculation was applied to the system, the organic matter removal improved to 77,6-96,7% while that of NNH_4^+ spiked to 70,4%. Recirculation offers improves the dilution of the incoming influent, thereby enhancing better contact between the contaminants in the fresh WW and the biomass.

7. USEFUL CONSIDERATIONS AND CONCLUSIONS

- After years of studies and implementations, the scientific community and indeed the larger society especially those in the rural areas, have widely recognized that CWs when incorporated into the wastewater treatment process, is a reliable treatment technology for wastewater especially now that very stringent legislations limit components of wastewater discharge into the environment, and more importantly, CWs could be used to treat wastewater in large quantity. The advances in the design and operation of CWs accomplished over the years have significantly increased pollutant removal efficiency, particularly nitrogen and organic material, and the sustainable application of this treatment system has also been significantly improved.

- One of the factors that influence the efficiency of sub-surface CW in the removal of contaminants contained in WW is CW macrophytes. Although it is not the major removal mechanism route for nitrogen, organic matter and other contaminants, it is very important to place close attention to the appropriate macrophyte's species to be selected, the climatic conditions of the site where the sub-surface CWs are going to be sited, the strength of the WW to be treated, the composition of the effluent (which is most often determined by the stringent limit discharge imposed by various environmental laws), adaptive features of the macrophytes to saturation condition (which more often than not takes place), the growth potential of the macrophytes roots, the capacity to exude oxygen and carbon, the capacity of withstand high contaminants concentration. Only few plants meet up with these wide criteria something which explains the use of very few plant species in CW, examples of macrophytes used are *Typha spp. (Typhaceae), Phragmites spp. (Poaceae), Iris spp. (Iridaceae), Scirpus spp. (Cyperaceae), Juncus spp. (Juncaceae), and Eleocharis spp. (Spikerush)*, all these are emergent plants. The commonly used submerged plants are *Hydrilla verticillata, Vallisneria natans, Ceratophyllum demersum, Myriophyllum verticillatum,* and *Potamogeton crispus*. Other types are floating-leaved plants are *Nymphoides peltata, Nymphaea tetragona, Trapa bispinosa,* and *Marsilea quadrifolia*. The free-floating plants are *Eichhornia crassipes, Salvinia natans, Hydrocharis dubia* and *Lemna minor*. The use of macrophytes of the *same species* have been found not to significantly enhance contaminants removal efficiency in sub-surface CWs but when macrophytes of *different species* were employed, it was demonstrated to significantly enhance contaminants removal efficiency.

- One of the draw backs of the use of sub-surface CWs system at least in the past decades was its low removal efficiency of contaminants. This is sometimes due to clogging which increases saturation and indirectly limit the diffusion of oxygen deep to the deepest zone of the environment and the end-result is low efficiency. To prevent this, it is very important for fresh WW to pass through the preliminary stage of filtration, screening, to remove solids, especially inorganic solid matter of considerable sizes.

- The review also indicates that despite the improved nitrogen removal performances due to wetland macrophytes, classical nitrification and denitrification routes are still considered to be the major mechanism of nitrogen removal from wastewater especially in subsurface flow wetland systems. Most research-works have demonstrated that nitrogen removal by macrophytes is very low when compared to the removal mechanism of the denitrification. Also, aerobic biodegradation is faster than anaerobic.

- One of the drawbacks of the use of CWs is the large area demand for its construction as well as serving as breeding ground for mosquitoes. With very good design the constructed wetlands could be designed as a single *"reactor"* in which all the various biological and chemical processes would take place in compartments that could be referred to as *"cells"* in such a way that no one inhibits the others, all the processes occurring sequentially or in combination that way the whole configuration occupies less space. Biological control could be adopted by employing larvivorous fishes, *Gambusia affinis*, *Poecilia reticulata, Gasterosteus aculeatus, etc* … which feed on mosquito larvae.

- A major problem for optimizing the classical nitrogen removal route is the necessity of maintaining alkalinity of wastewater within an adequate pH range while guaranteeing sequential aerobic-anaerobic conditions. Single-stage sub-surface constructed wetlands cannot achieve high removal efficiency of total nitrogen, TN, due to their inability to provide both aerobic and anaerobic conditions at the same time. Vertical-flow constructed wetlands provide good condition to successfully remove ammonia-N but very limited denitrification takes place in these systems. On the other hand, horizontal flow constructed wetlands provide good conditions for denitrification but the ability of these system to nitrify ammonia is very limited. Therefore, various types of constructed wetlands may be combined in what is known as the hybrid systems with each other in order to exploit maximally the specific advantages of the individual system. It would have been innovatively efficient to have a CW where aerobic and anaerobic conditions take place sequentially and in an adequate order, in such a way that the pH of the wastewater being treated can be regulated and controlled. This definitely demands extensive research that would focus in this area in the future.

- One of the problems of the use of sub-surface CWs in the treatment of WW, especially when it is of high strength, is that the removal efficiency of N, BOD and COD is lower in autumn and winter but robust in spring and summer. This could be explained by the fact that most of the biological processes that take place in sub-surface CWs are catalysed by enzymes secreted by micro-organisms that are predominantly bacteria in nature. Temperature affects microbial activities for example denitrification, a biological process carried out by heterotrophic facultative bacteria that takes place at a temperature range of 15-35°C but its optimum temperature range is taken to be between 25-27°C but at a very low temperature it proceeds very slowly. Nitrification, a bio-chemical process that precedes denitrification for it to take place, is also affected by temperature, its optimum temperature range is 30-40°C and again at a temperature of 5-6°C, occurs very slowly. Apart from temperature, some biological processes need oxygen to function, dissolved oxygen in sub-surface CWs like in other aqueous medium is affected by salinity, pressure and of course by temperature. At higher temperature, the level of dissolved oxygen decreases and at very temperature, it does increase. The problem is that at low temperature, photosynthesis rate proceeds very slowly, very little microbial decomposition takes place which directly slow down atmospheric contact for diffusion, making the dissolved oxygen level to be low. The end-result of the consequence of low temperature is that sub-surface CW operates with very low removal efficiency of contaminants, since a major removal route, the denitrification rate is proceeding at a very slow pace. A way out of this problem is to use internal CW liners that not only serve to keep the WW in the system and prevents ground water from entering the system but also one that serves as *an insulant* that keeps the temperature of the CW system within a specified range, most liners are made from polyvinyl chloride (PVC)

- To guarantee a CW system with a very high efficiency, as previously noted above, various types of CW have been combined to form different configurations, but it must be stressed that for the configuration to operate at a very high efficiency, it must be in line with the theory that governs the various processes that lead to the removal of contaminants, especially nitrogen and organic matter. The configuration that has been found to give the best removal efficiency is the combination of HF-SSCW and VF-SSCW. In my opinion, the best global configuration would be one in which there is a series of HF-SSCW; the number depends on the strength of the wastewater.

- Apart from putting in place the best configuration to guarantee a paramount efficiency, some operations have also been found to enhance it, like the use of different substrates in VF-SSCW to obtain a tidal flow, increasing the recircle ratio, the use of by-pass when one takes into cognisance that it takes a HRT of between 0-2,3 days for the removal of BOD_5 that is hoping that there is a low quantity of difficult bio-degradable organic matter and a HRT of between 2-10 days for the removal of N, the adoption of the wet and Dry technique in VF-SSCW to promote aeration, employment of forced aeration in HF-SSCW, and of course increasing the number of stages. The use of by-pass is extremely important because the denitrification process needs a carbon source for it to take place.

- It has been shown that the addition of external carbon to the sub-surface CW has increased efficiency, examples of substance added as external carbon source include ethanol, acetic acid, glucose etc. It Is also possible to add carbon internally, examples of internally carbon source include the use of wood mulch, rice musk, peat, zeolite, compost, slag, aluminium sludge,etc with wetland substrates.

- The newly Anammox and CANON processes offer a lot of advantages with significant potential for improved nitrogen removal efficiency. However, more research is needed to investigate and explore implanting this process in constructed wetlands.

- Further research is needed to produce a predominant microbial species and hydrophytes that would have a specific gene that can sequentially target nitrogen removal, and if possible, a specified organic compound, using biogenetical techniques through gene modification, in order to improve process performance.

7. CONSIDERACIONES ÚTILES Y CONCLUSIONES

- Después de años de estudios e implementaciones, la comunidad científica, la sociedad en general y las comunidades rurales en particular, han demostrado que el uso de humedales construidos en el proceso de tratamiento de aguas residuales resulta particularmente adecuado hoy en día, por las estrictas legislaciones actuales que limitan los niveles de contaminantes que se pueden verter en aguas residuales y, además, por la posibilidad de tratamiento de volúmenes elevados. Los avances en el diseño y la operación de estos humedales han mejorado significativamente la eficiencia de la eliminación de contaminantes, en particular nitrógeno y materia orgánica, así como la sostenibilidad del tratamiento.

- Uno de los factores que claramente afecta al rendimiento de los humedales construidos subsuperficiales es el tipo de macrofitos empleados. Aunque no representan el principal mecanismo de eliminación de nitrógeno, materia orgánica y otros contaminantes, es importante la adecuada selección de las especies a emplear, las condiciones climáticas del terreno donde se ubicará el sistema de humedales, la carga de aguas residuales a tratar, la composición del efluente (normalmente limitada por las correspondientes leyes medioambientales), la adaptabilidad de los macrofitos a las condiciones de saturación (situación que suele ocurrir), el potencial de crecimiento de sus raíces, la capacidad de exudar oxígeno y carbono o la capacidad de resistir altas concentraciones de contaminantes. Son pocas las plantas que cumplen todos estos requisitos, lo que explica el uso de pocas especies de macrofitos; *Typha spp. (Typhaceae), Phragmites spp. (Poaceae), Iris spp. (Iridaceae), Scirpus spp. (Cyperaceae), Juncus spp. (Juncaceae), y Eleocharis spp. (Spikerush)*, constituyen ejemplos de plantas emergentes. *Hydrilla verticillata, Vallisneria natans, Ceratophyllum demersum, Myriophyllum verticillatum,* and *Potamogeton crispus* son ejemplos de plantas sumergidas, mientras que *Nymphoides peltata, Nymphaea tetragona, Trapa bispinosa,* and *Marsilea quadrifolia* representan ejemplos de plantas de hojas flotantes, mientras que *Eichhornia crassipes, Salvinia natans, Hydrocharis dubia* and *Lemna minor* son ejemplos de plantas flotantes libres. Se ha encontrado que el uso de macrofitos de la misma especie no aumenta significativamente el rendimiento de los humedales construidos subsuperficiales, pero sí cuando se usan diferentes especies.

- Una de las principales desventajas del uso de humedales construidos subsuperficiales, al menos en décadas anteriores, ha sido el bajo rendimiento en la eliminación de contaminantes, debido, en ocasiones, a su obstrucción, lo que aumenta la saturación e, indirectamente, limita la difusión de oxígeno a la zona más profunda. Para evitar esto, es importante someter las aguas residuales a una etapa preliminar de filtración, cribado, para eliminar sólidos, sobre todo aquellos de materia inorgánica de tamaño considerable.

- La revisión también pone de manifiesto que, a pesar de la mejora en la eficacia de la eliminación de nitrógeno por el uso de macrofitos, la ruta clásica de nitrificación y desnitrificación sigue siendo el principal mecanismo de eliminación de nitrógeno en humedales construidos subsuperficiales. Además, se han demostrado que la biodegradación aeróbica es más rápida que la de anaeróbica.

- Otras desventajas de los humedales construidos son la necesidad de una gran superficie para su construcción, así como favorecer el crecimiento de mosquitos. Un bueno diseño de un humedal construido podría realizarse con un único "*reactor"* en el que todos los procesos biológicos y químicos tendrán lugar en compartimentos, que puedes denominarse *"celdas"*, de tal modo que ningún proceso inhibe a otro, todos ellos ocurren secuencialmente o combinados, pudiendo así lograrse una configuración global que ocupe menos espacio. El control biológico podría llevarse a cabo empleando peces larvívoros, como *Gambusia affinis*, *Poecilia reticulata o Gasterosteus aculeatus,* que se alimentan de las larvas de mosquitos.

- Un problema importante en la optimización de la ruta clásica de eliminación de nitrógeno es la necesidad de mantener la alcalinidad de las aguas residuales dentro de un intervalo de pH adecuado, que garantice las condiciones secuenciales aeróbicas-anóxicas. Los humedales construidos subsuperficiales de una etapa no son capaces de alcanzar altos rendimientos de eliminación de nitrógeno total, debido a su incapacidad de tener condiciones aeróbicas y anaeróbicas al mismo tiempo. Los humedales construidos de flujo vertical son capaces de eliminar eficazmente amonio-N, pero el proceso de desnitrificación es muy limitado. Por el contrario, en los humedales construidos de flujo horizontal se dan las condiciones adecuadas para la desnitrificación, pero su capacidad de nitrificación de amonio es muy limitada. Consecuentemente, se pueden combinan varios tipos de humedales construidos, dando lugar a sistemas híbridos, que permitan maximizar las principales ventajas de cada sistema. Constituiría una gran innovación, un sistema de humedales construidos en los que las condiciones aeróbicas y anaeróbicas se sucediesen secuencialmente en el orden correcto, de tal modo que el pH de las aguas residuales podría controlarse y modificarse. Este es un tema que necesitará ser estudiado muy extensamente.

- Otro problema detectado en el uso de humedales construidos, sobre todo en el caso de aguas residuales con cargas elevadas, es el bajo rendimiento de la eliminación de N, DBOD y DCO en otoño e invierno, que sube significativamente en primavera y verano. Este hecho puede explicarse considerando que la mayoría de los procesos biológicos que tienen lugar en estos humedales están catalizados por enzimas de microrganismos, de naturaleza predominantemente bacteriana. La temperatura afecta a la actividad microbiana, por ejemplo, en el caso de la desnitrificación, un proceso biológico llevado a cabo por bacterias facultativas heterotróficas en un intervalo de temperaturas de 15-35°C, aunque la actividad óptima ocurre entre 25-27° y procede muy lentamente a muy baja temperatura. La nitrificación, proceso bioquímico que debe preceder al proceso de desnitrificación, también se ve claramente afectada por la temperatura; su intervalo óptimo es de 30-40°C, procediendo de forma muy lenta a una temperatura de 5-6°C. Además de la influencia de la temperatura, muchos procesos biológicos necesitan oxígeno para que puedan tener lugar; el oxígeno disuelto en humedales construidos subsuperficiales, al igual que en otros muchos medios acuosos, se ve afectado por la salinidad, presión y, por supuesto, la temperatura. A alta temperatura, el nivel de oxígeno disuelto baja mientras que, este nivel sube a baja temperatura, pero, en estas condiciones, la fotosíntesis sucede muy lentamente, existe poca descomposición microbiana, lo que ralentiza el contacto atmosférico por difusión, haciendo que el nivel de oxígeno disuelto disminuya. El resultado a largo plazo, como consecuencia de bajas temperaturas, es que los humedales construidos subsuperficiales muestran bajos rendimientos de eliminación de contaminantes ya que la desnitrificación, una de las principales rutas de eliminación, ocurre muy lentamente. Una solución es usar revestimientos internos que no solo sirven para evitar que el agua del subsuelo entre al sistema, sino que también sirven como aislantes para mantener la temperatura de los humedales dentro de un rango específico. Muchos de estos revestimientos están hechos de cloruro polivinilo (PVC).

- Tal y como se mencionó previamente, para garantizar altos rendimientos se combinan varios tipos de humedales construidos en distintas configuraciones, lo que debe estar en línea con la teoría que rige los diversos procesos que conducen a la eliminación de los contaminantes, especialmente nitrógeno y materia orgánica. La configuración que ha dado una mejor eficiencia de eliminación es la que combina humedales horizontales y verticales. En mi opinión, la mejor configuración global sería la que combinase una serie de humedales horizontales acoplados con una serie de humedales verticales en paralelo; en ambos casos, el numero dependería de la carga de aguas residuales.

- Para garantizar rendimientos máximos de eliminación, además del diseño de la mejor configuración de los humedales, se ha encontrado que ciertas operaciones los mejoran, tales como el uso de diferentes sustratos en humedales construidos verticales para conseguir flujos de marea, el aumento de la relación de recirculación, el uso de by-pass cuando se tiene en cuenta que se necesita un tiempo de retención hidráulico de entre 0-2,3 días para la eliminación de DBO_5 y un tiempo de retención hidráulico de entre 2 y 10 días para la eliminación de nitrógeno, la utilización de la técnica húmedo y seco en humedales verticales para promover la aireación, el uso de aireación forzada en humedales horizontales y, por supuesto, el aumento del número de etapas. El uso de by-pass es muy importante porque el proceso de desnitrificación necesita una fuente de carbono para que pueda tener lugar.

- Diversos estudios han mostrado que la adición externa de carbono a humedales construidos subsuperficiales aumenta su rendimiento, ejemplo de fuente de carbono son etanol, ácido acético, glucosa, etc. También es posible añadir carbono internamente con los sustratos de los humedales; posibles fuentes son el mantillo de madera, almizcle de arroz, turba, zeolita, compost, escoria, lodos de aluminio, etc. con los sustratos de los humedales.

- Los recientes procesos de ANAMMOX y CANON ofrecen muchas ventajas, con un significativo potencial de mejora del rendimiento de la eliminación de nitrógeno. Sin embargo, aún faltan más estudios para implementar estos procesos en humedales construidos.

- Son necesarios más estudios referidos a la producción de especies microbianas e hidrofitos predominantes que tengan un gen específico que puedan tener como objetivo la eliminación de nitrógeno secuencialmente, y si es posible también, la eliminación de un compuesto orgánico específico, usando técnicas biogenéticas a través la de modificación genética, con el fin de mejorar la eficacia de proceso.

7. CONSIDERACIÓNS ÚTILES E CONCLUSIÓNS

- Despois de nos de estudos e implementacións, a comunidade científica, a sociedae en xeral e as comunidades rurais en particular, demostraron que o uso de humedais construidos para o tratamento de augas residuais resulta particularmente axeitato hoxe en día, pola estrita lexislación que limita os niveis de contaminantes que se poden verter. Os avances no deseño e a operación destos humidais teñen mellorado significativamente a súa eficacia na eliminación de contaminantes, en particular nitróxeno e materia orgánica.

- Un dos factores que claramente afecta ao rendimento dos humidais construidos subsuperficiais é o tipo de macrófitos empregados. Aínda que non representan o principal mecanismo de eliminación de nitróxeno, materia orgánica e outros contaminantes, é importante a axeitada selección das especies a empregar, as condicións climáticas do terreno onde se ubicará o sistema de humidais, a carga de augas residuais a tratar, a composición do efluente (normalmente limitada polas correspondentes leis medioambientais), a adaptabilidade dos macrófitos ás condicións de saturación, o potencial de crecemento das raíces, a capaciddade de exudar osíxeno e carbono ou a capacidade de resistir altas concentracións de contaminantes. Son poucas as plantas que cumpren todos estes requisitos, o que explica o uso de poucas especies de macrófitos; *Typha spp. (Typhaceae), Phragmites spp. (Poaceae), Iris spp. (Iridaceae), Scirpus spp. (Cyperaceae), Juncus spp. (Juncaceae), e Eleocharis spp. (Spikerush)*, constituen exemplos de plantas emerxentes. *Hydrilla verticillata, Vallisneria natans, Ceratophyllum demersum, Myriophyllum verticillatum,* e *Potamogeton crispus* son exemplos de plantas mergulladas, mentres que *Nymphoides peltata, Nymphaea tetragona, Trapa bispinosa,* e *Marsilea quadrifolia* representan exemplos de plantas de folla flotantes, e *Eichhornia crassipes, Salvinia natans, Hydrocharis dubia* e *Lemna minor* son exemplos de plantas flotantes libres. Atopouse que o uso de macrófitos da mesma especie non aumenta significactivamente o rendemento dos humedais construidos subsuperficiais, pero sí cando se usan diferentes especies.

- Unha das principais desvantaxes do uso de humedais construidos subsuperficiais, ao menos en décadas anteriores, foi o baixo rendemento na eliminación de contaminantes, debido en ocasións á colmataxe, o que aumenta a saturación e limita a difusión de osíxeno ás zonas máis profundas. Para evitar isto, é importante someter ás augas residuais a unha etapa preliminar de filtración, cribado, para eliminar sólidos, sobre todo aqueles de tamaño considerable.

- A revisión tamén pon de manifestó que, a pesares da mellora na eficacia da eliminación de nitróxeno polo uso de macrófitos, a ruta clásica de nitrificación e desnitrificación segue sendo o principal mecanismo de eliminación de nitróxeno en humedais construidos subsuperficiais. Asemade, se ten demostrado que a biodegradación aeróbia é máis rápida que a anaerobia.

- Outras das desvantaxes dos humedais construidos son a necesidade dunha gran superficie para a súa construcción, así como a presencia de mosquitos. Un bo deseño do humidal construido pode realizarse de forma que todos os procesos biolóxicos e químicos terán lugar secuencialmente ou combinados, poidendo así acadarse unha onfiguración global que ocupe menos espacio. O control biolóxico podería levarse a cabo empregando peixes larvívoros, como *Gambusia affinis*, *Poecilia reticulata ou Gasterosteus aculeatus,* que se alimentan das larvas de mosquitos.

- Un problema importante na optimización da ruta clásica de eliminación de nitróxeno é a necesidade de manter a alcalinidade das augas residuais dentro dun intervalo de pH axeitado, que garanta as condicións secuenciais aeróbica-anóxicas. Os humedais construidos subsuperficiais dunha etapa non son capaces de acadar altos rendimentos de eliminación de nitróxeno total, debido a súa incapacidade de ter condicións aerobias e anaerobias ao mesmo tempo. Os humedais construidos de flujo vertical son capaces de eliminar eficazmente nitróxeno amonicacal, mais o proceso de desnitrificación é moi limitado. Pola contra, nos humidais de fluxo horizontal danse as condicións axeitadas para a desnitrificación, mais a súa capacidade de nitrificación de amonio é moi limitada. En consecuencia, poden combianrse varios tipos de humedais constriudos, dando lugar a sistema híbridos, que permitan maximizar as principais vantaxes de cada sistema. Constituiría unha gran innovación un sistema de humidadis nos que as condicions aeróbicas e anaeróbicas tivesen lugar secuencialmente, de tal modo que o pH das augas poidese controlarse e modificarse. Este tema precisa ser estudado de forma máis extensa.

- Outro problema detectado no uso de HC, sobre todo no caso de augas residuais con cargas elevadas, é o baixo rendemento na eliminación de nitróxeno e materia orgánica en outono e inverno, aumentando na primavera e no verán. Este feito pode explicarse considerando que a mayoría dos procesos biolóxicos que teñen lugar en estes humedais están catalizados por enzimas de microorganismos, principalmente bacterias. A temperatura afecta á actividade microbiana, por exemplo no caso da desnitrificación, un proceso biolóxico levado a cabo por bacterias facultativas heterótrofas nun intervalo de temperaturas de 15-35ºC, aínda que a actividade óptima ocurre entre 25-27ºC e procede moi lentamente a moi baixas temperaturas. A nitrificación, proceso bioquímico que debe precederao proceso de desnitrificación, tamén se ve claramente afectada pola temperatura, o seu intervalo óptimo é de 30-40ºC, procedendo de forma moi lenta a unha temperatura de 5-6ºC. Asemade da influencia da temperatura, moitos procesos biolóxicos precisan osíxeno para que poidan ter lugar, o osíxeno disolto en humidais construidos subsuperficiais, vese afectado pola salinidade, presión e temperatura. A alta temperatura, o nivel de osíxeno disolto baixa, mentras que sube a baixas temperaturas, mais nestas condicións a fotosíntese sucede moi lentamente, existe pouca descomposición microbiana, o que ralentiza o contacto atmosférico por difusión, facendo que o nivel de osíxeno disolto diminúa. O resultado a longo prazo, como consecuencia das baixas temperaturas, é que o HC subsuperficiais amosan baixos rendimentos de eliminación de contaminantes xa que a desnitrificación, unha das pricipais rutas de eliminación, ocurre moi lentamente. Unha solución é usar revestimentos internos que non só sirven para evitar que a auga do subsolo entre no sistema, senon que tamén sirven como illantes para manter a temperatura dos humidais dentro dun rango específico. Moitos destes revestimentos están feitos de PVC.

- Tal e como se mencionó previamente, para garantir altos rendementos combínanse varios tipos de HC en diferentes configuracións, o que debe estar en liña coa teoría que rixe os diversos procesos que conducen á eliminación dos contaminantes, especialmente nitróxeno e materia orgánica. A configuración que ten acadado unha mellor eficiencia de eliminación é a que combina humedais horizontais e verticais. Na miña opinión, a mellor configuración global sería a que combinase unha serie de humidais horzontais acoplados cunha serie de humidais verticais en paralelo. En ambos casos o número dependería da carga das augas residuais.

- Para garantir rendimentos máximos de eliminación, ademáis do deseño da mellor configuración dos humidais, atópase que certas operacións os melloran, tais como o uso de diferentes sustratos en HC verticais para acadar flujos en marea, o aumento da relación de recirculación, o uso de by-pass cando se ten en conta que se precisa un tempo de retención hidráulico de entre 0-2,3 días para a eliminación de DBO_5 e un tempo de retención hidráulico de entre 2 e 10 días para a eliminación de nitróxeno, a utilización da técnica húmido e seco en humidais verticais para promover a aireación, o uso de aireación forzada en humidais horzontais e, por suposto, o aumento do número de etapas. O uso de by-pass é moi importante porque o proceso de desnitrificación precisa unha fonte de carbono para que poida ter lugar.

- Diversos estudos teñen demostrado que a adición externa de carbono a humedais construidos subsuperficiais aumenta o seu rendemento. Exemplo de fontes de carbono son etanol, ácido acético, glucosa, etc. Tamén é posible aumentar o carbono dos sustratos dos humidais engadindo mantillo de madeira, almizcle de arroz, turba, zeolita, compost, escoria, lodos de aluminio, etc.

- Os recentes procesos de ANAMMOX e CANON ofrecen moitas vantaxes, cun significativo potencial de mellora do rendemento na eliminación de nitróxeno. Porén, aínda faltan máis estudos para implementar estes procesos en humidais construidos.

- Son precisos máis estudos refereridos á producción de especies microbianas e hidrófitos predominantes que teñan un xen específico que poida ter como obxectivo a eliminación de nitróxeno secuencialmente, e se é posible tamén, a eliminación dun composto orgánico específico, usando técnicas bioxenéticas a través da modificación xenética, coa fin de mellorar a eficacia do proceso.

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