Addis Ababa University
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School of Graduate Studies
Department of Chemical Engineering

Dairy Wastewater Treatment Using Horizontal Subsurface Flow Constructed Wetland Planted With *Tyhpa Latifolia* and *Scirpus Lacustris*

By

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A Thesis Submitted to the School of Graduate Studies of Addis Ababa Institute of Technology, in Partial Fulfillment of the Requirements for the Degree of Master of Science in Chemical Engineering (Environmental Engineering)

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Abstract

Environmental pollution resulting from the discharge of untreated industrial effluents has been putting serious risks on the survivability and perpetuation of all living things including human beings for the past few years. From the various treatment methods natural means of treating industrial wastewater have been exercised in many parts of the world for they are environmentally permissible and help to keep its natural values. Among those natural ways of treating wastewater constructed wetlands have been utilized for the past few decades. In this research three horizontal subsurface flow constructed wetlands (HSSFCWs) have been used for the treatment of dairy wastewater discharged from Ada milk factory. The first HSSFCW was kept as control with only gravel and the rest two HSSFCWs were planted with widely used emergent plant species, Typha latifolia and Scirpus lacustris. The wetlands flow rate was adjusted to 25 L/d and as hydraulic retention time (HRT) 8 days was used for three replicates of the experiments. After laboratory investigations were carried out it was found that the wetland planted with Scirpus lacustris showed better capacity in removing pollutants from the dairy wastewater than the wetland planted with Typha latifolia. The average removal efficiencies of the wetland planted with Scirpus lacustris and the unplanted wetland were found as follows, respectively. BOD (84.2%, 65.2%), COD (84.4%, 68.2%), TN (43.4%, 22.8%), NH_4^+-N (84.2%, 59.2%), NO_3^-N (97.5%,88.7%), TSS (65%,70%,),VSS(32%,43%), TP (53.8%,27%), OP (80.7%,75.2%), FOG (95.3%,93%), TCa (39%,21.8%) and TC (89.7%,69.3%). Besides their importance in wastewater treatment, these emergent plants have versatile uses among these their medicinal and dietary values were widely recorded. Thus, it is environmentally advisable mechanism to utilize constructed wetlands for wastewater treatment. From laboratory investigations it was found that BOD, TN, NH_4^+-N, NO_3^--N, TP and PO_4^{3-}-P are the main factors for the pollution of the Dairy wastewater.

Keywords: **Constructed wetland, *Scirpuslacustris, Typhalatifolia,* dairy wastewater**

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List of Abbreviations

ADP	Adenosine Di-Phosphate
AF	Anaerobic Filter
AFOs	Animal Feeding Operations
ANOVA	Analysis of Variance
ATP	Adenosine Tri-Phosphate
BOD	Biochemical Oxygen Demand
CAFOs	Confined, Animal-Feeding Operations
CDPHE-WQCD	Colorado Department of Public Health and Environment Water Quality Control Division
CFCs	Chlorofloro Carbons
CIP	Cleaning in Place
COD	Chemical Oxygen Demand
CW	Constructed Wetland
DAF	Dissolved Air Floatation
DO	Dissolved Oxygen
Ec	Electrocoagulation
EC	Electrical Conductivity
EPA	Environmental Protection Agency
FOG	Fat, Oil and Grease
FWS	Free Water Surface
HRT	Hydraulic Retention Time
HSSFCWs	Horizontal Subsurface Flow Constructed Wetlands
IF	Impeller Flotation

LSD	Least Significant Difference
MCL	Maximum Contaminant Level
MF	Membrane Filter
MPN	Most Probable Number
NBOD	Nitrogenous Biochemical Demand
NF	Nanofiltration
PAM	Polyacrylamide
PH	Power of Hydrogen
RO	Reverse Osmosis
SBR	Sequential Batch Reactor
SSF	Subsurface Flow
SSFCWS	Subsurface Flow Constructed Wetlands
TC	Total Coliform
TDS	Total Dissolved Solids
TN	Total Nitrogen
TP	Total Phosphorous
TSS	Total Suspended Solids
UASB	Upflow Anaerobic Sludge Blanket
USDA	United States Department of Agriculture
USEPA	The U.S. Environmental Protection Agency
VFAs	Volatile Fatty Acids
VSS	Volatile Suspended Solid

1. Introduction

1.1. Background

Wastewaters from agricultural animal operations contribute large quantities of sediment, biochemical oxygen demand and nutrients to receiving waters. Dairy milking wastewater is an example of a dominant agricultural source of nonpoint source pollution [1]. In modern societies the proper management of these wastewaters is a necessity and not an option. Environmental authorities require, through current standards, that wastewaters receive adequate treatment to meet effluent quality standards. These standards often times approach that of freshwater streams. Therefore, it is necessary to develop both inexpensive and sustainable waste management practices with low energy input requirements. A more recent best management approach for purification of wastewaters is the use of constructed wetlands. The constructed wetlands (CWs) are considered as low-cost alternatives for treating municipal, industrial and agricultural wastewater. Those naturalized treatment systems also have been demonstrated to have significant potentials for both wastewater treatment and resource recovery. Therefore, CWs have been frequently used for nutrient removal from polluted rivers and lakes. Wetland filtration systems, for example, could reduce up to 30-67% total phosphorous (TP) and 30-52% total nitrogen (TN) of the hypereutrophic lake water. Compared with conventional treatment systems, CWs, which are of low cost, easily operated and maintained, can be potentially applied in developing countries with serious water pollution problems. [2]

However, due to lack of awareness, these systems have not been widely used. To treat polluted water, it is practically urgent to consider new, cheap and environmentally friendly approaches. To this end, constructed wetlands might be effective in treating nutrient pollution as well as in restoration of river ecosystems. We noted that plant uptake and storage were both important factors responsible for nitrogen and phosphorous removal in the three types of CWs. Milk production in the last decades has been modified from rural and local activity to more industrialized one resulting in serious environmental pollution. The presence of the macrophytes enhances several functions in the CW system: assisting solid sedimentation, reducing algae production, providing surface area for microbial growth, improving nutrient uptake, and releasing oxygen [3].

Dairy wastewater can be a major contributor to the cultural eutrophication of surface waters. Preliminary results are promising when wetlands are a component of a farm-wide waste management plan, but they are ineffective without pre-treatment of the wastewater. The feasibility of constructed wetlands varies with waste characteristics and climate. Further, as urban areas encroach on rural land; greater demands are placed on water resources. Nutrient and solids removal in wetlands is facilitated by shallow water which maximizes the sediment to water interface, high primary productivity, the presence of aerobic sediments and anaerobic sediments, and the accumulation of litter [4]. Nitrogen enters a dairy wastewater treatment wetland in either an organic or inorganic form. The inorganic forms are nitrate (NO_3^-), nitrite (NO_2^-), ammonia (NH_3), and ammonium (NH_4^+). Ammonia may be lost from the system through volatilization, taken up by plants or microbes, or oxidized to nitrate in the nitrification process. Similarly, ammonium may be taken up in the biota or nitrified. In addition, because of its positive charge, it can be sorbed onto negatively charged soil particles. Nitrate and nitrite are removed from the water by plant uptake or denitrification [5]. Once nitrogen has been denitrified; it is released to the atmosphere as nitrous oxide (N_2O) or dinitrogen gas (N_2).

Denitrification brings about a removal of nitrogen from the aqueous system and it is the most important removal pathway for nitrogen in most wetlands. As the organic nitrogen is mineralized, it enters the inorganic nitrogen cycle. High nitrogen removal rates have been noted in many wetland studies [6]. Because the transformations of nitrogen involve microbial processes, nitrogen removal is enhanced during the growing season. Bio-available forms of phosphorous may be taken up by plants or sorbed onto soil particles. Adsorption of phosphorus is the main mechanism for phosphorus removal in wetlands. Because phosphorus adsorption is enhanced under aerobic conditions, the removal of phosphorus in the anaerobic conditions of wetlands may be less than on dry soil [4].

1.2. Statement of the Problem:

The discharge of wastewater to the environment without any treatment poses significant risk on the public health, environment and it also decreases the access to clean water. Industrial wastes leads to contamination of water, soil and air when they are discharged without being subject to treatment or when they are treated using inappropriate methods [7].Ethiopia holds large potential for dairy development due to its large livestock population, the favorable climate for improved, high-yielding animal breeds, and the relatively disease-free environment for livestock [8].Dairy factories in Ethiopia are increasing in numbers among these: Ada, Mama, Lame, Family, Modjo milk factories are some of the known dairy factories in Ethiopia. Nowadays, in Ethiopia, the production of milk has been transformed from local scale to industrial scale, discharging huge amount of wastewater to the surroundings reluctant. Like other factories, Ada milk factory, has had problems ever since it was established in its wastewater removal techniques that discharges odorous wastewater full of organic matter, suspended solids, nitrogen and phosphorous into the living environment causing serious health problems to animals and human beings. This condition has the potential to minimize the quality of water bodies by increasing the organic matter load [9].Live algal mats can dramatically alter the dissolved oxygen concentration in the benthos and water column which increases during the day and decreases at night. After the algae dies, excessive organic matter results in biological oxygen demand (BOD). Increasing organic carbon availability and reducing oxygen concentrations creates conditions facilitating mercury methylation both in stream and in downstream reservoirs [10].

Excessive amounts of live algae can also cause wide daytime swings in pH due to the uptake of carbonic acid (a source of carbon dioxide) for photosynthesis. Although large mats and long filaments are signs of "eutrophication" (nutrient enrichment) of waters, moderate growth of this alga can occur in high quality water [11]. Dairy effluent generally contains high concentrations of nutrients, particularly N, P and K [12]. The fate and movement of dairy effluent derived phosphorous has become an important issue in many countries, because effluent phosphorous is commonly accumulated in soils, and can enter waterways through runoff and contribute to surface water eutrophication [13],[14].

Similarly, when dairy effluent application is based on Nitrogen loading, over-supply of other nutrients such as potassium may cause soil nutrient imbalance and animal health problems [15]. Odor is a public nuisance commonly associated with handling and application of dairy effluent that needs to be addressed [16]. Dairy effluent contains many pathogens that can potentially contaminate the water supplies of animals and humans [17]. The application of dairy wastes to land has been an accepted disposal method for many years but there are few studies investigating the long-term effects of dairy effluent irrigation on soils. The effect of casein wastewater applied to pasture for 15 years was studied [18]. The studies found marked increases in inorganic and organic forms of nitrogen and phosphorus in the soil, soil bulk density had decreased, and the soil had higher rates of respiration and more earthworms.

Further, due to turbidity and color photosynthesis may be restricted there-by affecting the primary link in the food chain [12]. Dairy effluent contains variable but often significant levels of potassium (typically as salts), and can be used to supply pasture or crop potassium requirements [19]. Significant potassium loadings may be applied to paddocks in dairy effluent, but although grazing and fodder conservation can export significant amounts of potassium, soil potassium levels can still become very high [20]. Excessive quantities of potassium in soils can lead to animal health problems, soil nutrient imbalances and detrimental environmental impacts [21]. Excessive quantities of potassium in soils can lead to potassium losses off site through leaching into groundwater or surface runoff. Although potassium is largely retained in soils, it can leach from coarse, sandy, well drained soils, where loadings are excessive or through bypass flow mechanisms [22]. As potassium can occur in the soil as a salt, high soil potassium salt contents can contribute to soil salinity, leading to reduced pasture production on highly saline soils [23]. Excessive supply of potassium with effluent application can result in luxury potassium uptake by pasture, which can considerably increase potassium intake by animals. The high potassium concentration in pasture has the effect of suppressing the uptake of other cations such as Calcium and Magnesium [24]. When Magnesium concentration in animal serum is lowered to a critical level, it can increase risk of "grass staggers" or hypomagnesaemia, a Magnesium deficiency disorder that reduces milk production and creates health problems varying from milk stress and tremors to sudden death [21]. Odor is a public nuisance associated with management and application of dairy effluent. Odorants in dairy wastes include ammonia, hydrogen sulfide, organic sulfur compounds, volatile fatty acids, and many other compounds [19]. Nitrogen losses of as high as50% or more of applied Nitrogen via ammonia volatilization are common following surface application of dairy effluent. In addition to the loss of nutritional value, ammonia volatilization and subsequent deposition can be a major source of pollution of waterways [25]. Ammonia volatilization not only causes Nitrogen loss, it can also result in odor problems [19].

1.3. Objectives of the Study:

1.3.1. General Objective:

The general objective of this study was to evaluate the performance of horizontal subsurface flow constructed wetland systems planted with *Typha Latifolia* and *Scirpus Lacustris* for the removal of pollutants from dairy wastewater

1.3.2. Specific Objectives:

The specific objectives of this study were:

- To characterize dairy wastewater forBOD_5, COD, TN, NO_2^--N,NO_3^--N, NH_4^+-N, TP, OP, TSS, TDS, EC, DO, Temperature, pH, VSS,FOG,TP,TC and Ca.
- To evaluate the removal efficiency of HSSFCW systems in the treatment of Dairy wastewater for:BOD_5, COD, TN, NO_2^--N,NO_3^--N, NH_4^+-N, TP, OP, TSS, TDS, EC, DO, Temperature, pH, VSS,FOG,TP,TC and Ca.
- To compare the removal efficiency of horizontal SSFCWs planted with the two emergent plants *Typha Latifolia* and *Scirpus Lacustris* for the parameters:BOD_5, COD, TN, NO_2^--N,NO_3^--N, NH_4^+-N,OP, TSS, TDS, EC, DO, Temperature, pH, VSS,FOG,TP,TC and Ca.

2. Literature Review

2.1. Dairy Wastewater Generation and Characteristics

Animal Feeding Operations (AFOs) have been widely recognized as a significant cause of surface water impairment, air pollution, and ground water contamination. Among many AFOs, dairy farms are the largest wastewater generators, contributing 48% of animal wastewater according to EPA surveys. According to EPA Feedlots Point Source Category Study, dairy farm wastewater has average chemical oxygen demand (COD) concentrations of 4997 mg/L and biochemical oxygen demand (BOD_5) of 1003 mg/L [26]. The COD concentration varies in the range of 2000–7000mg/L depending on wastewater management, climate, operation conditions, and types of flushing. The high COD concentration is due to waste milk (produced by washing milking equipment), detergent, manure, and waste feeds combined in the washing or flushing of holding pens and exit alleys. Dairy effluents contain dissolved sugars and proteins, fats, and possibly residues of additives. The key parameters are biochemical oxygen demand (BOD), with an average ranging from 0.8 to 2.5 kilograms per metric ton of milk in the untreated effluent; chemical oxygen demand (COD), which is normally about 1.5 times the BOD level; total suspended solids, at 100–1,000milligrams per liter (mg/l); total dissolved solids: phosphorus (10–100 mg/l), and nitrogen (about 6% of the BOD level). Cream, butter, cheese and whey production are major sources of BOD in wastewater [27]. The waste load equivalents of specific milk constituents are: 1 kg of milk fat = 3 kg COD; 1 kg of lactose = 1.13 kg COD; and 1 kg protein = 1.36 kg COD

Table 2.1. Product Losses in the Dairy Industry [65]

Operation	Product losses (%)		
	Milk	Fat	Whey
Butter/transport of skimmed milk	0.17	0.14	n.a.
Butter and skimmed milk powder	0.60	0.20	n.a.
Cheese	0.20	0.10	1.6
Cheese and whey Evaporation	0.20	0.10	2.2
Cheese and whey powder	0.20	0.10	2.3
Consumer milk	1.9	0.7	n.a.
Full-cream milk powder	0.64	0.22	n.a.

n.a. Not applicable.
Note: Data are expressed as the percentage of the volume of milk, fat, or whey processed

The wastewater may contain pathogens from contaminated materials or production processes. A dairy often generates odors and, in some cases, dust, which need to be controlled. Most of the solid wastes can be processed into other products and by products. Accordingly, 0.7–1.7 m^3 /ton of milk wastewater generate a BOD_5 of 500–1500 mg/L. According to studies of the waste characteristics of dairy farms, large-scale dairies produce a great deal of wastewater [28].

Table 2.2.Dairy Wastewater Characteristics [27]

Potential pollutant source	Biochemical Oxygen demand(ppm)	Nitrogen (ppm)	Phosphorous (ppm)	Volume gallons per 100 cows
Milking Centerwaste	400-10,000	80-900	25-170	73,000
Silage Leachate	12,000-90,000	4,400	500	105,000
Barnyard Runoff	1,000-10,000	50-2,500	5-500	80,000
Dairy Manure	20,000	5,600	900	660,000
Domestic Waste	150-250	150-250	5-10	365,000

2.2. Pollution Characteristics and Current Treatment Practices of Dairy Industry Waste

The average volume of wastewater in dairies is currently 1.3 l/kg milk. This results in considerable wastewater disposal costs. Hygiene is the most important factor in milk processing and the production of dairy products. This necessarily results in the use of considerable volumes of water for cleaning purposes. In addition, considerable quantities of wastewater with volatile milk constituents, fats and proteins occur when milk is being processed, particularly during evaporation and spray-drying. Pretreatment of effluents consists of screening, flow equalization, neutralization, and air flotation (to remove fats and solids); it is normally followed by biological treatment. If space is available, land treatment or pond systems are potential treatment methods. Other possible biological treatment systems include trickling filters, rotating biological contactors, and activated sludge treatment [28]. Pretreated dairy effluents can be discharged to a municipal sewerage system, if capacity exists, with the approval of the relevant authority. Odor control by ventilation and scrubbing may be required where cheese is stored or melted. Dust control at milk powder plants is provided by fabric filters. After aerobic or anaerobic biological treatment of dairy wastewaters, the residual sludge is sent through a clarifying decanter which efficiently dewaters the sludge before the clean water is recycled back into the process [29].

2.3. Dairy Factory Wastewater

Dairy factory wastewaters commonly contain milk, byproducts of processing operations, cleaning products and various additives that may be used in the factory. Bovine milk typically contains water (87%), fat (4%), protein (3.5%), lactose (4.7%) and ash (0.8%) [30].The fat content ranges from 3-5% with the major component being triglycerides (98%). Other fat components include phospholipids (0.5-1% of fat) and sterols (0.2 - 0.5% of fat) [31]. Depending on the diet of the cows, milk can also contain traces of other organic compounds, such as terpenes, that originate from plants [32].The composition of dairy factory wastewaters depends on the type of products manufactured and whether wastewater streams within the factory are segregated. Condensate collected from the evaporation of milk or whey is one of the cleanest wastewaters although it may contain volatile organic

Components and possibly liquid droplets of milk or whey entrained into the vapor stream from the evaporators [33].

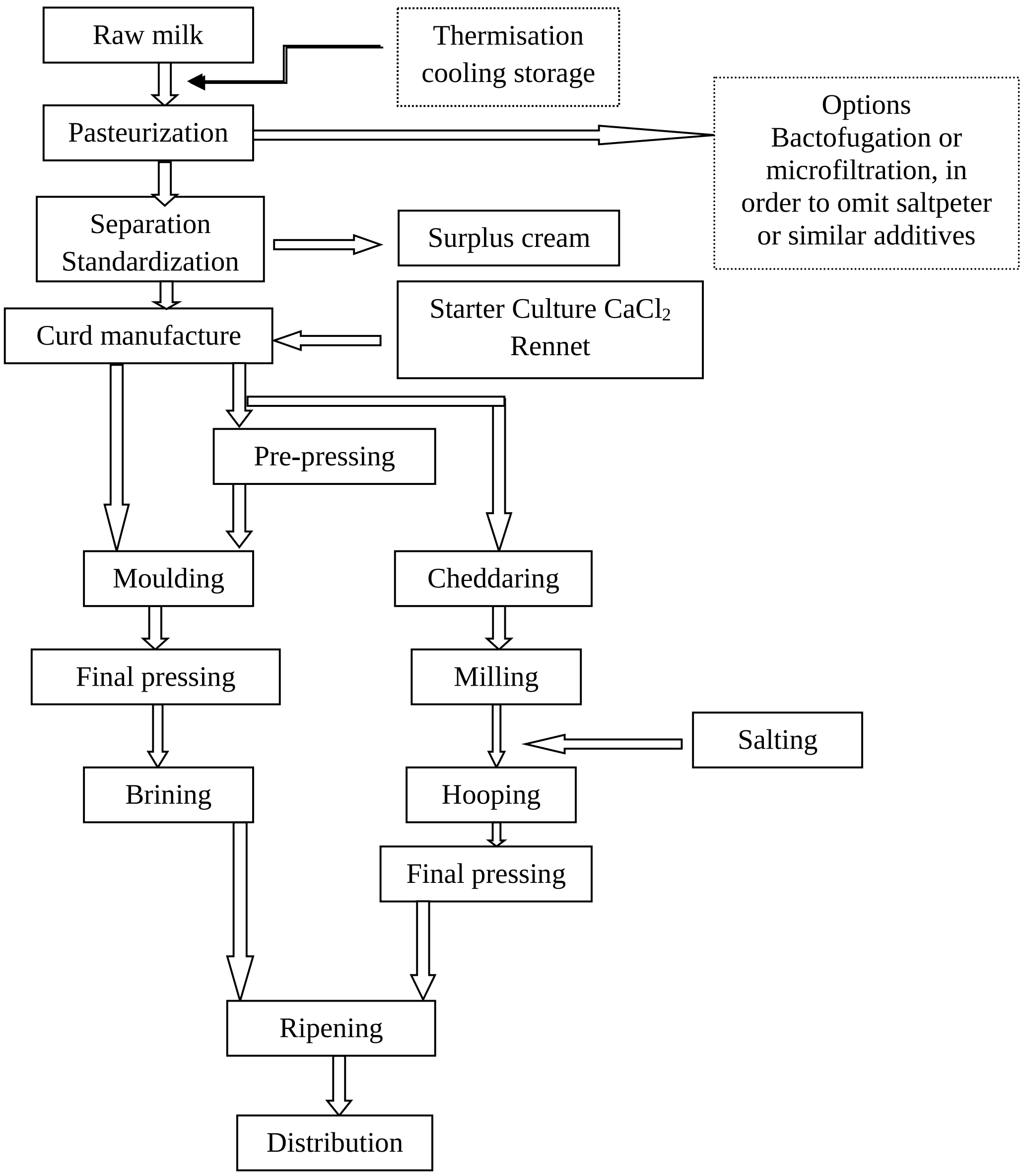

Figure 2.1: Flow diagram of dairy processing industry

Microbial contamination and cross contamination of products is a major issue for all dairy processing facilities and cleansing chemicals commonly contaminate wastewater. Sodium hydroxide is often used for removal of fats and proteins from milk lines and other surfaces. It contributes sodium to wastewater and increases wastewater pH. Nitric, phosphoric, hydrochloric, acetic and citric acids may also be used to remove remaining deposits, especially mineral scale. These acids can decrease the waste water pH significantly which can necessitate neutralization of the excess acid before treatment. Nitric and phosphoric acids contribute to the nutrient load of the wastewater and, as these nutrients can be difficult to remove from the wastewater, it can lead to accelerated eutrophication when discharged to the environment [33]. From an environmental standpoint, phosphoric acid is the least desirable acid to be used for cleaning so factories have moved away from using phosphoric acid to minimize discharge concentrations of phosphorus. Strong oxidants or bleaches such as peroxyacetic acid, sodium hypochlorite and chlorine dioxide are used for sanitizing equipment and chlorine bleaches may produce toxic organochlorine compounds that contaminate the wastewater. Other chemicals that are used for specific applications include enzymes and detergents that are particularly useful for cleaning cool surfaces and have less adverse downstream consequences. In most plants caustic chemicals are generally preferred for higher temperature surfaces which are more difficult to clean. Where wastewaters are ultimately applied to land, some factories use blends of sodium hydroxide and potassium hydroxide to minimize the concentration of sodium in the effluent. Considering that dairy factories produce a range of products and use a range of chemicals, it is not surprising that dairy processing wastewaters are highly variable in their composition. The frequency and stages of the cleaning process depend on the product runs, but occur at least daily [33]. A typical ‘cleaning in place’ (CIP) cycle begins with a water flush, followed by a sodium hydroxide wash, a water rinse, then an acid wash, followed by a second water rinse. A sanitizer may then be used. Murray Goulburn’s Koroit factory (with products including milk powders, butter and milk protein concentrate) uses neutral cleaners on all cold processing surfaces, and a recent upgrade to their CIP chemical recovery system has decreased sodium hydroxide usage by 45% with the result that the quantities of sodium discharged in the wastewater has been decreased. Various processes have been developed to minimize water use during cleaning. For example, ‘burst rinsing’ is a technique that uses less water than regular rinsing, and is suitable to pre-clean tanks and tankers [34]. The rinse water is pulsed in bursts rather than as a continuous stream. Burst rinsing has been introduced at Peters and Brownes (now Fonterra) in Balcatta, Perth where they produce flavored milk and ice cream. The burst rinsing technique takes longer to achieve the same level of cleanliness as conventional rinsing, but water consumption has been decreased by 15ML p.a. Water recycling is an alternative strategy for achieving water savings. Cleaning in place systems normally drain rinse waters and spent solutions to waste unless the systems have been designed to reuse or recover these streams. Chemical solutions can be reused and topped up with fresh chemicals until no longer effective, but a full recovery system typically uses membrane technology to recover cleaning chemicals and water [34]. The result is a decrease in water and chemicals used and discharged into the effluent. Condensate from the milk evaporation process is a relatively clean wastewater stream and has been reused for decades [35]. The recycled water is not suitable for all uses without further treatment but it is typically used for boiler feed water and membrane plant washing. Other water streams that are relatively clean that can be recycled include defrost water, boiler blow down, flushing water and pump sealing water [34].

Further purification of water streams may be achieved in different ways, but reverse osmosis allows reusing water (from milk washings) throughout the factory for processes such as cleaning flushes during the CIP cycles. Finally, effluent treatment creates sludge, and the sludge may be used as animal feed or for compost. Where possible waste streams can be treated to recover high-value products or bulk components to increase profits or offset disposal costs. Where this occurs the concentration of these products will often be low in the effluent stream. Whey, previously considered a waste product, is now sold in various forms and components such as protein concentrates and lactose, as well as some specialty ingredients including lactoperoxidase (an anti-microbial enzyme) and lactoferrin (anti-microbial protein) [34].

Table 2.3: Physico-chemical parameters of Dairy effluents [54]

S.No	Parameters	Untreated effluents	Treated effluent	I.S.I.Value
1	Colour	Whitish	Colourless	-
2	Temperature	28^0C	25^0C	-
3	pH	8.8	7.4	6.5-8.0
4	Dissolved Oxygen	Nil	3.5mg/l	4-6
5	Biochemical Oxygen Demand(BOD)	760mg/l	28.55mg/l	50
6	Chemical Oxygen Demand(COD)	1230mg/l	94mg/l	250
7	Total Dissolved Solids(TDS)	1000mg/l	480mg/l	1500
8	Total Solids(TS)	1310mg/l	560mg/l	1100
9	Total Suspended Solids(TSS)	310mg/l	80mg/l	Not above up to 450
10	Chlorides	630mg/l	90mg/l	600
11	Sulphate	395mg/l	75mg/l	Not above 1000
12	Fat,Oil and Grease	80mg/l	2.5mg/l	10

2.4. Dairy Wastewater Treatment Alternatives

Milking center wastewater is composed of excreta and bedding material washed from the parlor floor with high pressure hoses, waste milk, and milking equipment cleaning water that consists of detergents, acid and alkalizing agents, and sanitizers [36]. It was reported that the milk contributes a greater organic load due to milk contributing lactose, fats and proteins [37]. It was investigated by researchers that raw livestock wastewaters typically contain 2000-4000 mg/L BOD_5, 300-500 mg/LNH_3, and75-150 mg/L total Phosphorous [38]. Because milking center wastewaters consist of high concentrations of organic and nutrients in combination with less soluble fats, the treatment options can be limited. Numerous means of treating dairy wastes are suggested in the literature. Wide dispersal of milking center wastewater onto agricultural land is a common method of reducing its pollution potential. However, the dispersal practice may be too costly for some dairy operations due to the expenses associated with storage, transportation and spreading over a large area of land [39].

Furthermore, this practice may not be suited for all soils due to the composition of these wastes. The high levels of organic bio-solids may cause soil fouling in the land treatment areas [39].It was suggested that there was a possibility of dealing with solid manure wastes together with the milking parlor wastewater [40]. In this method the parlor wastewater is added to solid manure wastes to create a wastewater capable of being land applied by means of an irrigation system. This has the benefit of returning nutrients back to the soil. A drawback of the procedure is that the irrigation is not possible during winter months and therefore would require winter storage. Vegetative filter strip technology to dairy milk house wastewater produced at a Vermont farm [41]. Favorable results were obtained with retention of 95% total suspended solids, 89% total P, and 92% total Kjeldahl N on a mass basis. Despite the high reductions the effluent exceeded values expected for agricultural watersheds and effluent standards for wastewater treatment systems. Agricultural wastes have been applied to wetlands with reported successes. [42], [43].Interest in the treatment of dairy milk house wastewaters using primarily constructed wetlands has just more recently been investigated and reported. This study was conducted at the University of Connecticut's Storrs campus using three constructed wetlands in parallel [45]. Wetland vegetation used in the study utilized cattails (*Typha angustifolia*),common reed(*Phragmites australis*)and three-square bulrush (*Scirpus pungens*). In this study, they did not meet treatment objectives due to under sizing of the wetland. Their wetland sizing was based upon an assumed BOD_5 value, as found in an agriculture waste management handbook [44]. Another study that utilized constructed wetlands for the treatment of milk house wastewater was conducted between 1995 and 1997, in Frederick County, Maryland. In this study, the milk house wastewater was collected with barnyard runoff for treatment in two parallel wetland cells. The cells were planted with either cattails (*Typha latifolia*) or *Schoenoplectus tabernaemontani.* The Schoenoplectus later died and the cells were colonized by *Lemna minor L.*and *Echinochloa crus-galli.* They reported favorable results for all contaminants except for nitrate-nitrogen. One more study to be discussed later includes the study done by [46].In her study, she used the wetland constructed for this study along with a newly constructed lagoon to treat the milk house wastewaters. Her study provided a comparison of two technologies side by side treating wastewaters from the same source.

2.5. Wetlands

Wetlands are one of many aquatic systems where the major factor controlling the environment and the associated animal and plant life is water. By definition of the U.S. Fish and Wildlife Service, wetlands are those areas that exist between the upland and the aquatic environments. They consist of the transitional habitats where the water table is at or near the surface of the land and includes areas that have shallow water over land, up to a depth of 2 m [47]. The vegetation is dominated by hydrophytes, water loving plants, which can live in frequently saturated areas.

2.5.1. Types of Constructed Wetlands

Wetland is a general term that is often used when referring to three very different systems. This term often includes environments such as swamps, bogs, and marshes. They can be distinguished from one another by their specific hydrologic, nutrient and substrate conditions as well as the vegetation that occurs within each [27]. Vegetation offers the most obvious means of distinguishing between the different types of wetlands. Swamps are marked by the presence of vegetation in the form of water-tolerant trees, shrubs, and various other types of woody vegetation. Bogs on the other hand are wetlands dominated by acid-tolerant mosses that generally depend up on stable water levels that are acidic and low in nutrients [33].And then there are marshes with emergent vegetation such as cattail (*Typha*), bulrush (*Scirpus*),sedges (*Carex*),and reeds (*Phragmites*). Each of the wetland types, discussed above, has been used for wastewater treatment. Therefore, one of the first decisions to make when considering the use of constructed wetlands is the choice of the wetland type. This decision involves a consideration of the environmental conditions required to treat the given wastes as well as the feasibility of the wetland being successfully constructed and operated. From these standpoints, bogs and swamps often present more difficulty in constructed wetland application, as compared to marshes. Bogs dominated by mosses have been found to be hard to establish as well as difficult to operate for various pollutants.

Operational difficulties stem from the bog's limited adaptability to fluctuating water and nutrient loading levels [45]. In considering a swamp, its major setback is a required 5-20 years time period for development and full operational performance. On the contrary, marshes are better adapted to fluctuating water and nutrient levels as well as having greater tolerance of high pollutant concentrations. Furthermore, marshes have a great ability to inhabit a tremendous variety of soils, climatic and water quality conditions. Therefore, most constructed wetlands for wastewater treatment emulate marshes [33].When considering a marsh constructed wetland; there is also the choice of what type of hydraulic flow regime to use. The two types that are commonly used include the free water surface (FWS) and the subsurface flow (SSF) wetlands. In a FWS constructed wetland, the wastewater flows at a shallow depth through the emergent vegetation. Downward flow of water into the soil is prevented by the presence of an impermeable layer. This layer separates the wetland basin from the natural soil by the installation of an artificial barrier. With a subsurface flow constructed wetland the objective is to have the wastewater pass through the substrate of the wetland. These systems consist of substrate made up of a permeable medium, often sand and gravel, which allows the wastewater to flow laterally through the medium. One of the reported advantages of this system is the decreased potential of odors and mosquitoes due to subsurface flow [30].Construction costs for the two marsh systems are different. It was surveyed that 37 constructed wetlands was used and found that subsurface flow wetlands often have a higher construction cost due to the required permeable substrate acquisition and placement [35]. The average costs found were $55,000/ha ($22,000/ac) for FWS systems as opposed to the $215,000/ha ($87,000/ac) SF systems. However, when figured on a unit flow basis, the cost advantage went to the SF concept because of the smaller size required. Average cost reported for FWS systems was $206/m^3 ($0.78/gal) and for SF systems

$163/m^3 ($0.62/gal).In their survey of constructed wetlands, it was found that a significant number of SSF systems were experiencing surface flow [35]. The cause of this surface flow is perceived to be the clogging of the voids in the media with organic and/or inorganic material. If this clogging exists, the perceived advantages of a SSF concept are negated as one is left with a FWS wetland system. One of these requirements was that SF type systems were not to be used until research demonstrated their long-term effectiveness. Hence, in this study the term wetland will only refer to FWS systems. It was Listed that four major system components of a constructed wetland. This includes the plants, soils, bacteria, and animals. They go on to say that the function and the system performance, are influenced by water depth, temperature, pH, and dissolved oxygen concentration [30].

2.5.2. Components of Constructed Wetland

It was listed by many researchers that there are four major system components of a constructed wetland. This includes the plants, substrates, bacteria, and animals. They go on to say that the function and the system performance, are influenced by water depth, temperature, pH, and dissolved oxygen concentration [30].

➢ **Substrates, Sediments and Litter**

Substrates used to construct wetlands include soil, sand, gravel, rock, and organic materials such as compost. Sediments and litter then accumulate in the wetland because of the low water velocities and high productivity typical of wetlands. The substrates, sediments, and litter are important for several reasons:

- They support many of the living organisms in wetlands substrate permeability affects the movement of water through the wetland
- Many chemical and biological (especially microbial) transformations take place within the substrates
- Substrates provide storage for many contaminants the accumulation of litter increases the amount of organic matter in the wetland.

Organic matter provides sites for material exchange and microbial attachment, and is a source of carbon, the energy source that drives some of the important biological reactions in wetlands. The physical and chemical characteristics of soils and other substrates are altered when they are flooded. In a saturated substrate, water replaces the atmospheric gases in the pore spaces and microbial metabolism consumes the available oxygen. Both the physical and chemical attributes of soils can vary widely and are important to consider in construction and operation of constructed wetlands. Because of the saturated conditions of wetland substrate, it is dominated by anaerobic (reducing) conditions [37]. Since oxygen is consumed more rapidly than it can be replaced by diffusion from the atmosphere, substrates become anoxic (without oxygen). This reducing environment is important in the removal of pollutants such as nitrogen and metals.

- **Marsh Plants**

Wetland plants function in two important ways. The stems and leaves extending through the water column provide a large surface area for the attachment of microbial populations. Another function is in the way the plants transport gases to and from the roots and rhizomes in the wetland substrate. The wetland plants function in several ways to treat wastewater. Transferring of oxygen to the root zone plays a major role. Roots have been found to "leak" oxygen to the nearby soil thus creating a thin-film, aerobic region, in an otherwise anaerobic substrate, capable of supporting aerobic microbes. These aerobic microbial populations are capable of modifying trace organics, nutrients, and metallic ions. The by-products produced by the aerobic microbes are easily utilized by many of the anaerobic microbes located in the saturated soil, away from the aerobic thin film. The water column and plants work together, both above and below the water surface, to provide water treatment benefits [27]. Leaves and stalks above the water provide a canopy of shade, which functions to limit sunlight penetration. This shading tends to control alga growth by limiting light penetration to the water surface where they grow. If allowed to establish, the algae could deter oxygen transfer to the water column at the water-atmosphere interface [38]. Additionally, the shading effect of the plants has been found to decrease the water temperature in the summer [42]. Furthermore, the vegetation in the water column provides a substrate for attached microbial growth. The vegetation also provides a means of venting the gaseous byproducts of anaerobic decomposition in the subsoil via their internal aeration system or aerenchyma [42].

It was reported that wetland vegetation may retard water flows and in doing so cause reductions in suspended solids. It was further stated that the emergent vegetation reduces the water volume due to high transpiration rates of the plants [27]. In a survey of constructed wetlands, it was found that the most common types of vegetation used were cattail (*Typha*), bulrush (*Scirpus*) and giant reed (*Phragmites*) [35].It was also reported that *Phragmites* seemed to offer some treatment advantages over the other two; however it provided little or no habitat or other ecological benefits. Furthermore, their survey found that one third of the FWS systems used only cattails. These three commonly used constructed wetland plants have a tendency to create and/or maintain a single-species wetland environment by out-competing or inhibiting other plants. Selecting the correct plant will depend upon the climate and the goal of the wetland. The plant species selected must be capable of surviving the climatic conditions where the constructed wetland is to be used if yearly plantings are to be avoided [33]. If a goal of the constructed wetland is to remove nutrients one must consider the ability of the plant species to uptake and store the nutrients of interest. The storage ability of the plant relates to the amount of a particular nutrient that can be accumulated during a growing season, and therefore could be removed from the system if harvested. If frequent harvesting is to be considered, then the uptake capacity gives some indication as to the rate at which nutrients can be taken up by the plant, and at what frequency harvesting should be considered. It was also studied the storage and uptake capability of four common emergent macrophytes, and the results found for nitrogen and phosphorus are given in Table 2.4.

Table 2.4. Emergent Plants Nutrient Storage and Uptake. [39]

Emergent plants	Nitrogen		Phosphorous	
	Storage (kg/ha)	Uptake (kg/ha)	Storage (kg/ha)	Uptake (kg/ha)
Typhaspp (cattail)	250-1560	600-2630	45-375	75-403
Juncus (rush)	200-300	800	40	110
Scirpus (bulrush)	175-530	125	40-110	18
Phragmites (reed)	140-430	225	14-53	35

It was found that cattail (*Typha spp.*) had the greatest potential for storage and uptake of both nitrogen and phosphorus. Furthermore, the findings indicate more than 50% of nutrients were stored in belowground portions of the emergent plants. This was attributed to emergent plants having more below ground than above ground supportive tissue. The below ground plant tissue has a greater potential for storing the nutrients over a longer period. Therefore, there is a decreased necessity to conduct frequent or annual harvesting of plant matter in order to achieve nutrient removal. Both vascular plants (the higher plants) and non-vascular plants (algae) are important in constructed wetlands [45]. Photosynthesis by algae increases the dissolved oxygen content of the water which in turn affects nutrient and metal. Constructed wetlands attract waterfowl and wading birds, including mallards, green-winged teal, wood ducks, moorhens, reen and great blue herons, and bitterns. Snipe, red-winged blackbirds, marsh wrens, bank swallows, red tailed hawks, and Northern harriers feed and/or nest in wetlands.

- ***Typha latifolia***

Typha spp. (Cattails) (Typhaceae) are erect rhizomatous perennial plants with joint less stems. Rhizomes are extensive branched, produce aerial shoots at intervals and grow in shallow depth in horizontal direction. Leaves are flat to slightly rounded and obtain height up to 3 m. Inflorescence is a densely compact cylindrical, 15-50 cm long spike, which can produce up to 200,000 seeds with a high percentage of viability [6].

Figure 2.2: Roots and rhizomes of *Typha Latifolia* growing in HF constructed wetland

Cattail species are commonly found inhibiting shallow bays, irrigation ditches, lakes, ponds, rivers and both brackish and fresh water marshes. There are four major Typha species, among others, found in wetlands:*Typha latifolia L.* (Common cattail, Broad-leaved cattail),*Typha angustifolia* (Narrow-leaved cattail),*Typha domingensis* Pers. (Southerncattail, Santo Domingo cattail),*TyphaglaucaGodr*. (Blue cattail).*T.latifolia* and *T. angustifolia* are cosmopolitan species, *T. latifolia* is not found in central and south Africa. *T. glauca*, a hybrid of *T. latifolia* and *T.angustifolia* , is most common in North America and *T. domingesis* is foundin subtropical and tropical parts of Americas, Australia and Africa. The most common Typha species – *T. latifolia* and *T. angustifolia* could be distinguished according to the spike position. While in *T. latifolia* , there is no space between male and female spikes, in *T. angustifolia* both spikes are separated. These two species also differ in environmental requirements. *T. angustifolia* prefers water depth between 30 and 110 cm and it copes well with short-term decrease of water level and even drought. On the other hand, *T. latifolia* prefers water depth between 20 and 50 cm and does not like water level fluctuations. Typha plants are one of the most famous edible and useful plants in the world, often referred to as "supermarket of the marsh". The young leaves are excellent salad greens and can be cooked as potherbs before the pollen spike is ripe, it can be boiled and eaten much like corn on the cob [6]. However, at present the value as food source is generally negligible. Cattail leaves can be used for production of mats, chairs or baskets. Typha seeds could also be mixed with clay to produce bricks with good insulation properties [29].Typha is a very productive species, with maximum aboveground biomass values found in natural stands exceeding 5,000 g DMm-2. In constructed wetlands, Typha plants are usually used in free water surface systems where maximum above ground biomass is comparable with natural stands [6]. The use of Typha in HF constructed wetlands is limited mainly because the underground structures (roots and rhizomes) are very shallow, sediments occupied by Typha are usually more anaerobic then in the presence of other plants and also the rate of humification, i.e. creation of soil layer within the root zone, is much faster as compared to other species.

- ***Scirpus lacustris* (*Schoenoplectusacutus,Scirpusacutus, Schoenoplectuslacustri*s):**

It is called tule, common tule, hard stem tule, tule rush, hard stem bulrush, or viscid bulrush, is a giant species of sedge in the plant family Cyperaceae, native to freshwater marshes all over North America. The common name derives from the indigenous Mexican word *tullin* (Nahuatl=bulrush), and was first applied by the early settlers from New Spain who recognized the marsh plants in the Central Valley of California as similar to those in the marshes around Mexico City[79].It has a thick, rounded green stem growing to 1 to 3 metres (3–10 ft) tall, with long, grass like leaves, and radially symmetrical, clustered pale brownish flowers. Tules at shorelines play an important ecological role, helping to buffer against wind and water forces, thereby allowing the establishment of other types of plants and reducing erosion. Tules are sometimes cleared from waterways using herbicides. When erosion occurs, tule rhizomes are replanted in strategic areas.

Figure 2.3: Roots of *Scirpus Lacustris* species

- **Marsh Microbes**

The component of the wetland system contributing considerably to the decomposition of pollutants comes from the wetland microorganisms. Some of the groups of microbes noted include bacteria, fungi, algae, and protozoa. Contaminants are altered by these organisms to obtain nutrients or energy to carry out their life cycles. Seeding or transplanting of these organisms in constructed wetlands is generally not necessary as they are ubiquitous and naturally occurring in most waters [33].A fundamental characteristic of wetlands is that their functions are largely regulated by Microorganisms and their metabolism [40]. Microorganisms include bacteria, yeasts, Fungi, protozoa, rind algae. The microbial biomass is a major sink for organic carbon and many nutrients. Microbial activity:

- transforms a great number of organic and inorganic substances into innocuous or insoluble substances
- alters the reduction/oxidation (redox) conditions of the substrate and thus affects the processing capacity of the wetland
- involved in the recycling of nutrients

Some microbial transformations are aerobic while others are anaerobic and many bacterial species are facultative anaerobes. Microbial populations adjust to changes in the water delivered to them. Populations of microbes can expand quickly when presented with suitable energy-containing materials. When environmental conditions are no longer suitable, many microorganisms become dormant and can remain dormant for years [10].The microbial community of a constructed wetland can be affected by toxic substances, such as pesticides and heavy metals, and care must be taken to prevent such chemicals from being introduced at damaging concentrations.

- **Water**

Wetlands are likely to form where land forms direct surface water to shallow basins and where a relatively impermeable subsurface layer prevents the surface water from seeping into the ground. A wetland can be built almost anywhere in the landscape by shaping the land surface to collect surface water and by sealing the basin to retain the water. Hydrology is the most important design factor in constructed wetlands because it links all of the functions in a wetland and it is often the primary factor in the success or failure of a constructed wetland [35]. While the hydrology of constructed wetlands is not greatly different than that of other surface and near-surface waters, it does differ in several important respects:

- Small changes in hydrology can have fairly significant effects on a wetland and its treatment effectiveness
- Because of the large surface area of the water and its shallow depth, a wetland system interacts strongly with the atmosphere through rainfall and evapotranspiration. Density of vegetation of a wetland strongly affects its hydrology, first, by obstructing flow paths as the water finds its sinuous way through the network of stems, leaves, roots, and rhizomes and, second, by blocking exposure to wind and sun [35].

- **Animals**

Constructed wetlands provide habitat for a rich diversity of invertebrates and vertebrates. Invertebrate animals, such as insects and worms contribute to the treatment process by fragmenting detritus and consuming organic matter. The larvae of many insects are aquatic and consume significant amounts of material during their larval stages, which may last for several years. Invertebrates also fill a number of ecological roles; for instance, dragonfly nymphs are important predators of mosquito larvae. Although invertebrates are the most important animals as far as water quality improvement is concerned, constructed wetlands also attract a variety of amphibians, turtles, birds, and mammals [27], [42].

2.6. Pollutant presence and removal in Constructed wetlands

Various nutrients are required by the biological organisms and plants of a natural aquatic system to survive. These same nutrients, if present in high levels, produce unstable environmental health conditions. Therefore, the treatment mechanisms within constructed wetlands can play an important role in wastewater treatment. The two prevalent nutrients polluting water bodies by resulting in a condition called nutrient over enrichment are nitrogen and phosphorus. Other pollutants typically in need of removal from wastewaters, in order to protect the receiving aquatic systems, include the organic matter and solids [40]. These pollutants are removed in wetlands through a combination of physical, chemical and biological mechanisms.

Excessive organic material in a natural aquatic system is often detrimental to the system's stability. This is due to the fact that it is used as food by aquatic organisms. During the utilization of the organic matter, dissolved oxygen in the water column is consumed. This activity can limit the availability of dissolved oxygen for the larger aquatic organisms that require the dissolved oxygen for their survival. When measuring the oxygen demand put on water body by the organisms from organic matter consumption the standard test used is referred to as carbonaceous biochemical oxygen demand (BOD) [41]. The reduction of this BOD level is usually a requirement for wastewaters before they may be discharged to the environment. The BOD entering a constructed wetland will be both soluble and insoluble. Insoluble forms and settleable BOD may be quickly removed from the water column by the physical mechanisms of sedimentation and filtration. Once settled to the bottom, the solids may be stored until consumed by the aquatic organisms. Both insoluble and soluble BOD may be removed from the water by coming into contact with substrates covered with aquatic microorganisms. Within a wetland, the vegetation extending through the water column can produce significant amounts of surface area for these organisms to grow [42]. Findings showed that planted wetlands had greater removal of total BOD than unplanted wetlands, particularly when higher BOD loading rates were used in the wetlands. The COD levels are generally higher than the BOD as more compounds can be chemically oxidized than can be biologically oxidized. However, for many wastes it is possible to correlate the COD with BOD [38]. That is both measure the organic matter concentration in wastewaters by considering the amount of oxygen needed degrade the organic matter.

Total Solids (TS) in wastewaters is made up of both the suspended (TSS) and dissolved solids (TDS). The Total Suspended Solids (TSS) is composed of settleable matter, floating matter and colloidal matter [38].The organic portion is referred to as Volatile Suspended Solids (VSS), and its measure gives an indication of the amount of organic material in suspension and available for biodegradation. The presence of solids within the water column decreases the light penetration through the water column. This affects the aquatic organisms that require light for photosynthesis. The removal of TSS is primarily accomplished via settling. The process is accelerated by more quiescent conditions. Within a wetland system the quiescent conditions are enhanced by the presence of the emergent vegetation. In wetlands TSS removal rates were not a function of hydraulic or solids loading, but appeared to be related to the detention time. Dissolved solids removal in a wetland is accomplished by microorganisms adsorbing and/or absorbing and breaking them down to obtain energy [42].

Within dairy wastewaters, nitrogen can be found in a variety of forms because of the various oxidation states it can take. Its form may often change from one to another if subjected to different physical and biochemical conditions of the water. One form of nitrogen that is of particular interest is ammonia. At high concentrations it is toxic to many fish and other aquatic organisms. Additionally, since it can be oxidized further it can produce a significant oxygen demand on a body of water. Generally, nitrogen in dairy wastewaters are initially in complex organic compounds originating from milk waste, bedding material, and feces, as well as in the ammonium ion (NH_4^+) from urea. Organisms utilize the organic compounds, but are unable to oxidize the nitrogen from the organic material. Instead, the organic nitrogen is released as NH_4^+. The fate of NH_4^+; in aqueous environments is its utilization by nitrifying bacteria as a source of energy as shown in the nitrification equation-1 below.

During the nitrification process, oxygen in the water is utilized and creates what is called a nitrogenous biochemical oxygen demand (NBOD) [46].

Nitrification:

$NH_4^+ + 2O_2 \rightarrow NO_3^- + H_2O + 2H^+$ (2.1)

Denitrification:

$NO_3^- \rightarrow NO_2^- \rightarrow NO \rightarrow N_2O \rightarrow N_2$ (2.2)

The resulting nitrate (NO_3^-) produced during the nitrification process may be further broken down into various other nitrogen compounds during the denitrification process equation-2 being conducted by different aquatic organisms. During denitrification the nitrate is reduced eventually to one or more of the gaseous nitrogenous compounds shown. Each of these gaseous byproducts may be released to the atmosphere through volatilization and thereby removing the nitrogen from the wastewater. It has been found that several environmental conditions can affect the rate of one or more of these nitrogenous conversion reactions. One of the factors limiting the removal of nitrogen from the wastes is the opposite oxygen requirements of the nitrification and denitrification processes. As opposed to the nitrification process requiring oxygen, the denitrification process is suppressed by the presence of oxygen. The oxygen suppresses the enzymatic system of the denitrification organisms; therefore, this process often occurs most efficiently in anoxic areas [38]. The opposite requirements of the two processes are met within the benthic portion of the wetland due to the presence of the plant roots. It was reported that the roots of the aquatic plants leaked oxygen into the nearby benthic soils [42]. This creates an environment where aerobic and anaerobic environments are in proximity. Therefore, the aerobic nitrification byproducts may pass to the anoxic environment via a concentration gradient. In the anaerobic environment the denitrification organisms will produce the gaseous byproducts that are capable of passing through the aquatic environment and into the atmosphere and the rate of ammonification to be lower when hydrogen ion concentrations are outside the pH range of 6.5 to 8.5 [44]. They also reported that temperature has a large effect on ammonification rates as well. Ammonification rate constants were reported to double with a 10°C (18^{O}F) increase in temperature. In contrast, the temperature effect is not found to be highly inhibitory for the denitrification process. Laboratory studies showed that significant denitrification could occur at temperatures as low as 5^0C (41^0F), as well as within the 2-8°C (35.6-46.4°F) temperature range provided sufficient carbon was available and DO levels were below 2-3 mg/L [28]. Another means of nitrogen reduction in wetlands is through the aquatic vegetation. The aquatic vegetation takes up nitrogen and incorporates it into plant material, thereby suggesting the harvesting of the vegetation as a means of removing nitrogen. However, these same studies; also suggest that the harvesting would only account for 10% or less of the nitrogen removal for the wetland system [46], [29].

Phosphorus sources to water systems are primarily through its release from rocks containing phosphorus or through wastewater inputs. The process of phosphorus exchanging between the sediments and water is dependent upon several factors acting separately or in combination. Some of the more important factors include: lower dissolved oxygen levels causing phosphorus to be more soluble; water exchanges over or near the source, as it affects diffusion and transport; temperature, through its effect on microbial activity; pH; and the relative fractions of phosphorus in the sediment bound with inorganic and organic matter. Phosphorus is commonly the limiting nutrient in most aquatic systems. Therefore, when sufficient amounts of phosphorus are available to aquatic plants their growth becomes almost limitless. This extensive growth produces such conditions as increased plant matter buildup and dense algae mats, which ultimately lead to accelerated eutrophication and oxygen depletion of surface waters. Forms of phosphorus in the environment are numerous. Analysis of wastewater for phosphorus commonly includes orthophosphate (OP) and Total Phosphorus (TP). OP is a phosphorus analysis for the forms available for uptake, while TP analysis determines all sorbed and complexed inorganic and organic phosphorus. Removal of phosphorus in a wetland system occurs as a result of adsorption, complexation, and precipitation [47].

Subsurface flow constructed wetlands are effective systems for the removal of water pollutants and as such are widely used for the treatment of wastewaters in Europe [1]. The capacity of constructed wetlands to remove phosphorus is an issue that has not been satisfactorily solved. The Phosphorous removal potential of constructed wetlands is limited and is highly dependent on the nature of materials used for its construction, due to the Phosphorous saturation of media and consequently a reduction in the Phosphorous removal efficiency. Other factors that might affect the removal mechanisms include the growth of bio-film attached to the media, which reduces the contact and the interaction between the material and the treated water, and the inhomogeneous nature of media, which does not guarantee a consistent performance of the systems. To overcome the limitation of Phosphorous removal several alternatives have been suggested and tested. These include: (a) the construction of the whole system with chemically enriched media capable of removing Phosphorous; (b) chemical precipitation of Phosphorous at the pre-treatment stage of the system, and (c) the removal of Phosphorous in separate specific granular media filtration units. Previous experiments provided promising results, and a couple of materials showed good capacity to remove Phosphorous under laboratory conditions [59]. One of the materials in the trials showed a removal capacity of around 25 kg P m^{-3}. Previous studies show that the Phosphorous removal capacity of the media is associated with its chemical composition [6], specifically high content of Ca. Therefore, the materials selected for the tests included several sands and granular media known to have high Ca content.

The soil phosphorus cycle is fundamentally different from the N cycle. There are no valency changes during biotic assimilation of inorganic Phosphorous or during decomposition of organic Phosphorous by microorganisms. Soil Phosphorous primarily occurs in the +5 (oxidized) valency state, because all lower oxidation states are thermodynamically unstable and readily oxidize to $PO4^{3-}$ even in highly reduced wetland soils [6]. Phosphorus has only a minor gaseous phase (phosphine, PH_3). Phosphine is soluble in water, but has a high vapor pressure. It may be emitted from regions of extremely low redox potential, together with methane. Phosphorus exists mainly as apatites, with a basic formula $M_{10}(PO_4)_6X_2$.Commonly the mineral (M) is calcium, less often Al or Fe. The anion (X) is either F^-, Cl^-, OH^- or CO_3^{2-}, thus there exist fluor-, chloro-, hydroxy- and carbonate apatites. Diverse substitutions and combinations of (M) and (X) result in some 200 forms of Phosphorous occurring in nature [1]. Phosphorus does not show extensive biologically-induced fluxes to and from the atmosphere, as do carbon and nitrogen. Nor, in contrast to reduced forms of C and N, does it serve as a primary energy source for microbial oxidation. Nevertheless, soil organisms are intimately involved in the cycling of soil Phosphorous. They participate in the solubilization of inorganic Phosphorous and in the mineralization of organic Phosphorous.

Phosphorus in wetlands occurs as phosphate in organic and inorganic compounds,

that is as:

$$H_3PO_4 \Leftrightarrow H_2PO_4^- \Leftrightarrow HPO_4^{2-} \Leftrightarrow PO_4^{3-} \quad (2.3)$$

Orthophosphate occurs in ionic equilibrium, with $H_2PO_4^-$ and HPO_4^{2-} being the predominant species over pH range of 5 to9 [5]. Another group of inorganic phosphorus compounds are polyphosphates linearly condensed and cyclic. Organically bound phosphorus is present for example, in phospholipids, nucleic acids, nucleoproteins, phosphorylated sugars or organic condensed polyphosphates (coenzymes, ATP, ADP). Organic Phosphorous forms can be generally grouped into 1) easily decomposable Phosphorous (nucleicacids, phospholipids or sugar phosphates) and 2) slowly decomposable organic Phosphorous (inositol phosphates or phytin) [6].

Phosphorus transformations in soil and water column of wetlands are manifold and include: adsorption and desorption, precipitation and dissolution, plant and microbial uptake, fragmentation and leaching, mineralization, sedimentation and burial. Phosphorus that enters the wetland water column is rapidly absorbed by bacteria, periphyton, and plants. Radioisotope Phosphorous studies have shown that 10 to 20% of the Phosphorous is controlled by the biotic uptake initially [1]. Inorganic phosphorus transformations, subsequent complexes, and Phosphorous retention in wetland soils and sediments are controlled by the interaction of redox potential, pH values, Fe, Al, and Ca minerals, Organo-metallic complexes, organic matter content, clay minerals, hydraulic loading, and the amount of native soil Phosphorous [6].

2.7. New Technologies in the Dairy Industry Wastewater Treatment

The dairy industry involves processing raw milk into products such as consumer milk, butter, cheese, yogurt, condensed milk, dried milk (milk powder), and ice cream, using processes such as chilling, pasteurization, and homogenization. Typical by-products include buttermilk, whey, and their derivatives. The wastewaters discharged by raw milk quality control laboratories are more complex than the ones commonly generated by dairy factories because of the presence of certain chemicals such as sodium azide or chloramphenicol, which are used for preserving milk before analysis [2]. This section provides details on the latest developments and efforts in the dairy industry waste water treatment. Current Wastewater Treatment Process - Dairy Industry New Technologies in the Dairy Industry Waste Water Treatment are:

- Biomethanation of Whey and Cattle Dung
- Treatment of Dairy Industry Wastewater by Reverse Osmosis for Water Reuse
- Anaerobic Filter Reactor Performance for the Treatment of Complex DairyWastewater at Industrial Scale
- Influence of the Content in Fats and Proteins on the Anaerobic Biodegradability of Dairy Wastewaters
- Influence of Filtration Conditions on The Performance of Nanofiltration and Reverse Osmosis Membranes in Dairy Wastewater Treatment
- Anaerobic Treatment of Dairy Wastewaters: A Review
- Electrochemical Technologies in Wastewater Treatment
- Hydrolytic Enzymes as Coadjuvants in the Anaerobic Treatment of Dairy Wastewaters
- Effect of Enzymatic Hydrolysis on Anaerobic Treatment of Dairy Wastewater

- **Biomethanation of a Mixture of Salty Cheese Whey and Poultry Waste or Cattle Dung**

It aimed at improving the efficiency of anaerobic digestion of salty cheese whey in combination with poultry waste or cattle dung. Best results were obtained when salty cheese whey was mixed with poultry waste in the ratio of 7:3, or cattle dung in the ratio of 1:1, both on dry weight basis giving maximum gas production of 1.2 L/L of digester/d with enriched methane content of 64% and 1.3 L/L of digester/d having methane content of 63% respectively. Various conditions such as temperature and retention time have been optimized for maximum process performance [49].

- **Treatment of Dairy Industry Wastewater by Reverse Osmosis for Water Reuse**

The dairy industry is among the most polluting of the food industries in volume in regard to its large water consumption. It was related to investigations about practices of water management of 11 dairy plants. Treatment of the process water produced in the starting, equilibrating, stopping and rinsing processing units was proposed to produce water for reuse in the plant and to lower the effluent volume. Reverse osmosis of such wastewaters, collected in dairy plants, was performed after a prior check of their stability during storage. Filtration performances were focused on permeate flux versus water recovery and on water quality. Reverse osmosis water similar to available vapor condensates (produced in drying processes) can be achieved allowing this water to be reused for heating, cleaning and cooling purposes. A 540 m^2 RO unit is required to treat 100 m^3/d of wastewater with 95% water recovery [50].

- **Anaerobic Filter Reactor Performance for the Treatment of Complex Dairy Wastewater at Industrial Scale**

The wastewaters discharged by raw milk quality control laboratories are more complex than the ones commonly generated by dairy factories because of the presence of certain chemicals such as sodium azide or chloramphenicol, which are used for preserving milk before analysis. The treatment of these effluents has been carried out in a full-scale plant comprising a 12 m^3 anaerobic filter (AF) reactor and a 28 m^3 sequential batch reactor (SBR). After more than 2 years of operation, a successful anaerobic treatment of these effluents was achieved, without fat removal prior to the anaerobic reactor. The organic loading rates maintained in the AF reactor were 5–6 kg COD/m^3 /d, with COD removal being higher than 90%. No biomass washout was observed, and most of the fat contained in the wastewaters was successfully degraded. The addition of alkalinity is crucial for the maintenance of a proper buffer medium to ensure pH stability. The effluent of the AF reactor was successfully treated in the SBR reactor, and a final effluent with COD content below 200 mg/l and total nitrogen below 10 mg N/l was obtained [11].

- **Influence of the Content in Fats and Proteins on the Anaerobic Biodegradability of Dairy Wastewaters**

The relative amounts of fats, proteins and carbohydrates in wastewaters from dairy industries cause problems during their anaerobic treatment. The anaerobic biodegradability of two synthetic wastewaters, one rich in fats (chemical oxygen demand)(COD) ratio; Fats/Proteins/Carbohydrates: 1.7/0.57/1) and the other with a low fat content (COD ratio; Fats/Proteins/Carbohydrates: 0.05/0.54/1) was studied in samples with total COD ranging from 0.4 to 20 g/l. There were no problems of sludge flotation and the maximum biodegradability and methanisation were obtained when operating with wastewaters in the range of3–5 g COD/l. The intermediates of fat degradation (glycerol and long chain fatty acids) seemed not to reach concentrations high enough to affect the process. The anaerobic biodegradation of fat-rich wastes was slower than carbohydrate-rich wastes due to the slower hydrolytic step of fat degradation which prevented the accumulation of volatile fatty acids (VFAs) and favored the overall process. Carbohydrate-rich wastewater degradation produced free ammonia (FA) at concentrations near to inhibitory levels (62.2 mg FA/l), but in this case, ammonia production facilitated regulation of fall in pH caused by of the accumulation of VFA [2].

- **Influence of Filtration Conditions on the Performance of Nanofiltration and Reverse Osmosis Membranes in Dairy Wastewater Treatment**

Filtration performance and fouling of nanofiltration (NF) and reverse osmosis (RO) membranes in the treatment of dairy industry wastewater were investigated. Two series of experiments were performed. The first one involved a NF membrane for treating the chemical-biological treatment plant effluents. The second one used a RO membrane for treating the original effluents from the dairy industry. The permeate flux was higher at higher transmembrane pressures and higher feed flow rates. The curves of permeate flux exhibited a slower increase while the feed flow rate decreased and the pressure increased. Membrane fouling resulted in phermeate flux decline with increasing the feed COD concentration. Furthermore, the flux decline due to the COD increase was found higher at higher pressures for both NF and RO membranes [4].

- **Anaerobic Treatment of Dairy Wastewaters**:

Anaerobic treatment is often reported to be an effective method for treating dairy effluents. The main characteristics of industrial dairy waste streams are identified and the anaerobic degradation mechanisms of the primary constituents in dairy wastewaters, namely carbohydrates (mainly lactose), proteins and lipids are described. Primary attention is then focused on bench–pilot–full-scale anaerobic treatment efforts for dairy waste effluents. Combined (anaerobic–aerobic) treatment methods are also discussed. Finally, areas where further research and attention are required are identified [5].

- **Electrochemical Technologies in Wastewater Treatment**

This paper reviews the development, design and applications of electrochemical technologies in water and wastewater treatment. Particular focus was given to electro-deposition, electro- coagulation (EC), electro-flotation (EF) and electro-oxidation. Electro-deposition is effective in recover heavy metals from wastewater streams. It is considered as an established technology with possible further development in the improvement of space-time yield. EC has been in use for water production or wastewater treatment. It is finding more applications using either aluminum, iron or the hybrid Al/Fe electrodes. The separation of the flocculated sludge from the treated water can be accomplished by using

EF. The EF technology is effective in removing colloidal particles, oil & grease, as well as organic pollutants. It is proven to perform better than dissolved air flotation, sedimentation, impeller flotation (IF). The newly developed stable and active electrodes for oxygen evolution would definitely boost the adoption of this technology. Electro-oxidation is finding its application in wastewater treatment in combination with other technologies. It is effective in degrading the refractory pollutants on the surface of a few electrodes. Titanium-based boron-doped diamond film electrodes (Ti/BDD) show high activity and give reasonable stability. Its industrial application calls for the production of Ti/BDD anode in large size at reasonable cost and durability [55].

- **Hydrolytic Enzymes as Coadjuvants in the Anaerobic Treatment of Dairy Wastewaters**

An enzymatic extract produced by Penicillium restrictum having a high level of lipase activity (17.2 $U.g^{-1}$) was obtained by solid-state Fermentation using babassu cake as substrate. The enzymatic extract was used in the hydrolysis of a dairy wastewater with high fat contents (180, 450, 900 and 1,200$mg.L^{-1}$). Different hydrolysis conditions were tested, and it was determined that it should be carried out at a temperature of 35ºC, without agitation, with 10% v/v enzymatic extract and a hydrolysis time of 12 hours. Both crude and hydrolyzed effluents were then submitted to an anaerobic biological treatment. It was observed that for the enzymatically pretreated effluent there was a significant improvement in the efficiency of the anaerobic treatment. For the highest fat content tested (1,200 $mg.L^{-1}$), removal efficiencies of 19 and 80% were attained for crude and hydrolyzed effluents, respectively. In addition, a tenfold increase in the removal rate of COD from the hydrolyzed effluent (1.87 kg $COD.m^{-3}.d^{-1}$) was observed in relation to the crude effluent (0.18 kg $COD.m^{-3}.d^{-1}$). The results obtained in this study illustrate the viability of using a hybrid treatment (enzymatic-biological) for wastewaters having high fat contents [1].

- **Effect of Enzymatic Hydrolysis on Anaerobic Treatment of Dairy Wastewater**

The biological treatment of a synthetic dairy wastewater containing high levels of oil and grease (200, 600 and 1000 mg/l) was investigated, using two identical UASB reactors. One reactor was fed with wastewater from an upstream enzymatic hydrolysis step and the other with raw wastewater. The hydrolysis was carried out at 35 °C for 14 h, using an enzyme preparation obtained through solid-state fermentation, presenting pronounced lipase activity. The reactors were continuously operated with each fat concentration. The performance of both reactors was similar up to the concentration of 600 mg/l. However, the benefits of the hydrolysis step became evident with the highest concentration (1000 mg/l). COD removals averaged 90% in the reactor fed with the hydrolyzed effluent and 82% in the control reactor. The results showed that UASB reactors are able to operate, even when fed with high levels of oil and grease in dairy wastewaters [6].

2.8. Cold Weather Effects

In the application of constructed wetland technology, it is imperative that the climatic conditions be taken into account. Freezing temperatures in northern areas have presented problems with constructed wetlands treating animal wastes. It was found that their wetland cells froze solid during much of the winter, thus eliminating the microbial treatment of the wastewater [29]. However, others have found solutions to the ice problem. For example, a constructed marsh in Ontario Canada is used year round to treat municipal lagoon effluent [29]. In this wetland the short-circuiting caused by ice blockages was treated by raising the water levels to greater than 30 cm prior to the onset of winter. Freezing and short-circuiting aside the ice formation causes other problems. When freezing conditions produce ice formation that persists for more than a few days, the depletion of oxygen in the system is a possibility because of the reduction or prevention of surface re-aeration [30].

2.9. Periphyton (Attached Algae and Aquatic Plants) as Indicators of Water pollution:

Periphyton in streams and rivers are an important component of aquatic ecosystems, providing food for invertebrates, and thus fish, in local and downstream ecosystems [76]. Periphyton growth can be light-limited or nutrient-limited or both, and is influenced by temperature. In addition, periphyton communities can rapidly deplete waterways of nutrients, assuming no additional inputs, and communities vary compositionally (i.e., species types) with nutrient concentrations. Excessive periphyton growth can occur in rivers and lakes as a result of high water temperatures from reduced managed flows or excess nutrient production from human development on the landscape, through releases from wastewater treatment facilities, agricultural operations, deforestation, and soil disturbance, and therefore can serve as an ecological indicator for this disturbance. "Excessive growth" is defined here as growth that is not normal for the system and that causes local or downstream negative impacts such as changes in the particulate and dissolved organic carbon budget, nutrient cycling, biological and chemical oxygen demand, pH, and/or methylation and accumulation of mercury in fish. Increases in aquatic vegetation growth can change and negatively impact benthic macro invertebrate abundance and species richness and their functional role in the ecosystem as consumers of organic material and prey to larger invertebrates and vertebrates [76].

Figure 2.4: Periphytons in rivers

Periphyton community structure, species composition, and succession respond to environmental conditions and thus can be used to classify waterways [77]. In addition, these algal communities can and have been used as biotic indicators of ecological condition and change in condition in response to human and natural disturbance. Periphyton growth on the edge of Lake Tahoe has been suggested as a useful environmental indicator for human-induced nutrient enrichment in that system [77]. It is uncommon in waters low in calcium, nitrogen, and phosphorous. Moderate growth of this alga can occur in high quality water, though large mats and long filaments are signs of "eutrophication" (nutrient enrichment) of waters. Most freshwater algae are primarily growth-limited by the availability of phosphorous, and secondarily nitrogen. According to the U.S. Environmental Protection

Agency (EPA), periphyton is a complex assemblage of algae, cyanobacteria, micro-invertebrates, their secretions, and detritus attached to submerged surfaces. Most periphyton in the Everglades is considered calcareous due to abundance of the limestone (calcium carbonate) bedrock underlying the Everglades and from surface water inputs containing high cation concentrations. Periphyton is crucial and a fundamental part of the food web as the primary food source for small consumers, including fish and invertebrates. The South Florida Water Management District generated a model demonstrating the significance of periphyton for water quality and its interaction with other ecosystem components. Through physiochemical interactions with the surrounding environment and biota, periphyton affects many biological communities and ecosystem features including nutrient concentrations, soil quality, and dissolved gases. For example, periphyton provides dissolved oxygen through photosynthesis to sustain much of the aquatic life in its surroundings. Furthermore, because periphyton responds rapidly to changes in the environment, it is an indicator of changing conditions [76].

2.10. Air Pollution from Dairy Processing Industries:

In dairy plants air pollution is mainly caused because of the need for energy. In the process gasses may be discharged such as CO_2, CO, NO_x and SO_2.Table 2.5 gives the emissions into the air as a result of gas- and oil-combustion. No figures are available about the emissions into the air resulting from the use of electricity. Emissions of CFC’s and NH_3 into the air may come about as a result of leakage and stripping of chilling machines when out of use [78].

Table 2.5. Air emission of different gases from dairy processes [20]

Process	Air emission (kg/ton processed milk)	
Heating by burning gas or oil	CO	0.03
	CO_2	92
	NO_x	0.1
	SO_2	0.05
Producing milk powder	Fine dust	0.39
Cleaning	VOC	0.05

3. Materials and Methods

3.1. Materials and Equipments

3.1.1. Materials

Materials which are used to do the research work and used to analyze the parameters stated are the following:

- Emergent wetland plants- the selected wetland plants are *Typha latifolia* and *Scirpus lacustris* they are shown below in the figures

Figure.3.1: *Typha Latifolia* spp　　　Figure.3.2: *Scirpus Lacustris* spp

- Equalization tank (80L)-used to regulate the flow rate of the waste water into the wetland system by using a floating ball valve.
- Gravel size of 20mm-30mm diameter used in the treatment zone (middle) of the wetland. Gravel size of 40mm-80mm is used in the inlet and outlet zone of the wetland.
- Plastic bottles-are used to take samples for laboratory analysis.
- Plastic pipes-plastic pipes with 1 inch or 2.54 cm diameter made of polypropylene was used for the supply, distribution and collection of the wastewater.

- Controlling valve- a metal valve with 1-inch diameter was used to control the flow of the waste water entering into the wetland.
- Sedimentation tank (700L)-It is where suspended and settleable solids settle to the bottom of the tank.

3.1.2. Equipments

- BOD incubator (TS606/4)
- Drying oven (BSchickUNG-loading model100-800)
- BOD bottle, Erlenmeyer flasks (500mL), Aluminum weighing dishes, Glass-fiber filter disks, Suction flask, Membrane filter funnel, Gooch crucible, Filter paper(110mm), measuring cylinder (50mL and 100mL), weighing scale, evaporating dishes, desiccators, beaker(100mL)
- DO meter (HACH P/N HO30d LOVELAND. CO, USA)
- EC meter(Wagtech International N374,+M207/03 IM ,USA)
- PH meter (Wagtech N374, M128/03IM, USA)

3.2. Methods

Analytical reagents were used for the analysis of dairy wastewater composition and analysis was conducted for the samples taken from Ada milk factory, Bishoftu, Ethiopia. The wastewater was analyzed for COD, BOD_5, TN, NH_4^+-N, NO_3^--N, Ca, $NO2^-$-N, TP, PO_4^{3-}, FOG, TS, TC, VSS, TSS, TDS, DO, EC, Temperature and pH.:

3.2.1. Experimental Wetland System

The wetland system used for the research has three columns of horizontal SSFCWS with a dimension of 2m length, 0.65 m width and 0.6 m depth with surface area of 1.3 m^2. The bottom of the system has 1% slope from inlet to out let to achieve hydraulic head loss. This system is designed for an average waste water flow rate of 25 L/d (0.0025 m1/d) into each columns and HRT (hydraulic retention time) of 8 days. The void volume of the gravels was measured and found 0.227 m^3 (227L). Accordingly, the porosity of the gravel with diameter of 20mm-30mm was calculated by dividing the void volume to the total volume providing

0.35 or 35%. The empty volume of each SSFCW columns is 65L (0.65 m^3). The septic tank was used as a pre- treatment or as primary treatment unit which provided particle settling for two days. After the dairy waste water passed the primary treatment and the waste water then directly passed to a sixty liters equalization tank with a ball valve. Where the glow of the waste water is regulated before passing to the SSFCW columns. Hydraulic retention time of 6-8 days is recommended but for the dairy waste water has relatively higher solid loads and 8 days was used as HRT that provides more time for effective settling. To measure the inflow rate and the out flow rate of the waste water stop watch and graduated cylinder was used. The inflow rate was then found 17.4 ml/min and the out flow rate 14.8 ml/min. The media used in the SSFCW was gravel with diameter size of 20mm-30mm, the gravel was washed with water and it was refilled into the wetland cells to a height of 0.5m.The selected gravel size of 20mm-30mm was used based on the recommended gravel size range of 20mm-40mm for SSFCWS. The two emergent plants used for this research were *Typha Latifolia* and *Scirpus Lacustris*. The reason why these plants were selected among other plants including *Phragmites Australis* was that these plants have been prominent in treating polluted water. Specially, in treating dairy waste water which is full of Nitrogen and phosphorous *Typha Latifolia* and *Scirpus Lacustris* are advisable plant species. In addition to their use as bioremediation they have different purposes for instance; *Typha Latiflia* can be used as a source of food, medicine and for other uses. The rhizomes of *Typha Latifolia* are edible after cooking and removing the skin. Some cultures make use of the roots of *Typha Latifolia* as a poultice for boils, burns or wounds. The root-raw or cooked of *Scirpus Lacustris* which is rich in starch can be dried and ground into powder or made into syrup and this plant is a traditional medicine for cancer. After the gravel was filled into the wetland cells the plants were established on June 23, 2012. The first SSFCW cell was planted with *Typha Latifolia* and the second SSFCW cell was planted with *Scirpus Lacustris* and the third SSFCW cell was with only gravel used as a control. The plastic pipes used for the supply, distribution and collection of waste water are l inch (2.54 cm) diameter of polypropylene pipes. The sedimentation and equalization tank were connected through a single pipe. The two main components of the piping system in the SSFCW were the inlet and outlet pipes.

3.2.2. Sample Collection

Samples were collected from four different points of the SSFCWs at eight days interval a total of 13 samples were examined during the research experiments in three replicates. The first sample was taken from the settling tank and assigned X_1 and the second sample was taken after the waste water passed the sedimentation tank before entering the equalization tank and assigned X_2, the rest of the samples X_3, X_4 and X_5 were taken from the exit pipe of *Scirpus Lacustris*, *Typha Latifolia* and only gravel or control of the SSFCW, respectively. The samples were collected for one month starting September 12, 2012 to October 12, 2012 when the amount of rainfall is minimum and get good treatment performance of the SSFCW. After the plants reached acclimatization stage and fully grown the dairy wastewater was diluted with water at different percentages of 25%, 50%, 75% and 100% the samples were collected for eight days using plastic bottles.

3.2.3. Laboratory Analysis and Measurement

Almost all parameters were analyzed at JiJe labo glass laboratory and environmental protection Authority laboratory except DO and temperature for these parameters needs onsite measurement. The DO and thermometer were used to measure the dissolved oxygen and temperature, respectively. Measurement methods used are given in table 3.1.

Table 3.1.Laboratoy testing methods of some parameters

Parameters	Methods
Biochemical oxygen demand, BOD	5-day BOD Test
Chemical oxygen demand, COD	Open reflux method
Total solid,	Total solids dried at 103-105^{0}c
Total dissolved solids, TDS	Total dissolved solids dried at 180^{0}c
Total suspended solids, TSS	Total suspended solids dried at 103-105^{0}c
Volatile suspended solids, VSS	Fixed and Volatile solids ignited at 550^{0}c
Fat, oil and grease, FOG	Partition gravimetric method.
PH	Electrometric method
Electrical conductivity, EC	Laboratory method
Ortho phosphorous, OP	Vanadomolybdo phosphoric Acid colorimetric method.
Total phosphorous, TP	Vanadomolybdo phosphoric acid colorimetic method.
Total calcium, Ca	Direct Air- acetylene method
Total nitrogen, TN	Semi-micro kjeldahl method
Ammonium Nitrogen, NH_4-N	Titrimetric method
Nitrate nitrogen, NO_3-N	Ultraviolet spectro photometric method
Nitrite Nitrogen, NO_2-N	Colorimetric method

3.2.4. Statistical Analysis

Statistical analysis performed using JMPIN software. The data obtained then analyzed using one way Anova with 95% confidence level to compare the performance of each wetland cells removal of BOD_5, COD, TN, NH_4-N, NO_3-N, NO_2-N, TP, OP, TSS, TDS, PH, EC, VSS, Ca, and FOG, the statistical analysis was done using the mean, standard deviation, the analysis of variance (ANOVA) and measuring the least significant difference (LSD) at 5%. The removal efficiency of the SSFCWS for each waste water quality parameters was calculated using the following formula.

$$\text{Removal efficiency (\%)} = (C_i - C_e)/ C_i) \times 100 \quad (3.1)$$

where C_i= the concentration of the waste material in the influent, C_e = the concentration of the waste material in the effluent.

3.3. Site Description

Ada milk cooperative association is found in eastern Shewa zone in Oromia region in Ada woreda, Bishoftu Town. It is 47 kilometers in the east from Addis Ababa. Ada milk cooperative association is one of the milk factories which is working as cooperate organization. This cooperate organization was established in Liben woreda, Bishoftu Town in September 1998 with 34 founding members. The initial capital of the cooperation was only 3,400 ETB (400USD). Milk collection and marketing was started in January 2000, the amount of milk collected from founding members was only 308 liters per day or about 25,000 liters per month. Ada milk cooperate association has also recently installed modern machines capable of complexing, refrigerating and cooling 15,000 liters of milk per day. Over the past few years the organization has made a significant progress and its members grew to 787 and the number of dairy cows had become over 3,000 and the capital of the Organization has grown to 1,536,468 ETB in 2004 E.C.

4. Results and Discussions

4.1. Removal Performance of Sedimentation Tank (Pretreatment Unit)

After the dairy effluent was treated in the sedimentation unit for two consecutive days; September 03, 2012 and September 04, 2012 some pollutants showed promising decline in their concentration. In this research the mean measure of BOD_5 showed a decrease after pretreatment from 506±70 mg/l to 241.4±32 mg/l. This indicated the settling tank had average BOD_5 removal efficiency of 52.3%. The average influent BOD_5 value, that is, 241.4 mg/l indicated that the settling tank had a good performance in removing BOD even though the value did not comply with the international standard limit of 50 mg/l. The above result and laboratory analysis of BOD showed that most of the organic matter in dairy effluents was found as suspended solids of casein, lactose and Fat. Thus, the decrease in the organic matter concentration after settling was due to their settling with the solids found in the dairy waste water. The additional reason for the decrease in the BOD value was that aerobic bacteria which are important for organic matter degradation got sufficient aeration time or oxygen in the sedimentation tank and hence, minimizing the BOD value.

Figure.4.1: The wetland plants before feeding

Figure.4.2: The wetland plants after feeding wastewater

Accordingly, the mean COD concentration of raw wastewater was found to be 2520±189mg/l and the mean influent concentration of COD after settling was 359±53mg/l. This result indicated that the sedimentation tank had 85.7% average removal efficiency for COD. Since the concentration of COD after settling did not comply with the international standard limit of 250mg/l, the effluent was further treated by horizontal flow constructed wetlands to reach the standards. In here, the removal performance of the septic tank was high, even if there was sufficient aeration for organic matter oxidation, the attachment of the dairy organic matter with solids inhibited further oxidation of the organic matter.

The total solids concentration in waste effluent represents the colloidal form and dissolved species. In this research the TSS mean concentration decreased from 318±20mg/l as raw wastewater to 265±17mg/l as concentration after pretreatment. This result indicated that the settling tank had 17.1% removal efficiency for TSS .The average concentration of TSS after pretreatment i.e.265mg/l indicated that the value complies with the international standard value of not above 450mg/l. The volatile suspended solids (VSS) mean concentration decreased from 200±23mg/l as concentration of raw wastewater to 156±18mg/l as mean concentration after pretreatment. This result showed that the septic tank had 22% removal efficiency for VSS. Most of the solids in dairy wastewater are settle able but very small sized suspended solids needed more settling time to be effectively removed.

Fat, oil and grease influence waste water system and if present in excessive amount they might interfere with the aerobic and anaerobic biological processes and lead to decreased wastewater treatment efficiency. In this research the concentration of FOG in the raw wastewater was 406±29mg/l and after pretreatment the concentration of FOG was found to be 80±15mg/l. The result showed that the removal efficiency of the septic tank for FOG was 80.3%. From this study it was observed that the septic tank had good performance for removing FOG though the result does not agree with the international standard limit of 10mg/l. For most of FOG components are not found as solids i.e. they float over due to their lower density than water, it was difficult for the settling tank meet the standard limit in removing FOG. Thus, further treatment with the wetlands was required to meet the maximum allowable limit for discharging.

Nitrogen is limited in drinking water to protect the health of infants and may be limited in surface waters to prevent Eutrophication. Nitrogen can be removed in pond systems by plant or algal uptake, nitrification and denitrification and loss of ammonia gas to the atmosphere (evaporative stripping or volatilization). In this research the mean measured value of nitrogen as ammonium decreased from 4.35±2mg/l as raw concentration to 0.152±0.009mg/l as of influent concentration. This result indicated that the sedimentation tank had 96.5% removal efficiency. The decrease in the concentration of nitrogen as ammonium was due to the prevailing of most of the nitrogen in dairy effluents as ammonia that escapes the system by evaporation or volatilization. And in addition to, volatilization because of long time aeration some of the ammonia was converted to nitrate form hence increasing nitrate concentration and minimizing the amount of ammonia after pretreatment. The immediate decrease in the concentration of ammonia in the septic tank before entering to vegetated wetlands helped the survivability of the wetland plants for ammonia is very toxic gas for plants.

Among the parameters removed by sedimentation tank, nitrogen in nitrite form was the one with maximum efficiency. In this research study the concentration of nitrogen as nitrite decreased highly from 0.23±0.01mg/l to 0.002±0.0001mg/l after pretreatment. These results indicated that the sedimentation tank had 99.1% removal efficiency for nitrogen as nitrite. The decrease in the concentration of nitrogen as nitrite was due to the oxidation of nitrite into nitrate form by nitrifying bacteria. This was observed when the concentration of nitrogen as nitrate had increased highly after pretreatment. The maximum contaminant level (MCL, mg/l) of nitrogen as nitrite is 1mg/l and the concentration of nitrogen as nitrite after pretreatment was 0.002mg/l which is 1/500 of the MCL. Thus, the pretreatment unit was effective in removing nitrogen as nitrite which affects infants below the age of six months who drink water containing nitrite in excess of the MCL could become seriously ill and, if untreated, may die. Symptoms include shortness of breath and blue-baby syndrome.

Among the selected parameters nitrogen as nitrate showed an increase in concentration after pretreatment that is, from 0.58±0.15 mg/l as influent concentration to 15±6 mg/l as concentration after pretreatment. This result showed that after settling nitrogen as nitrate concentration had increased by 96.1%. This was due to the oxidation of ammonia and nitrite to nitrate form by nitrifying bacteria, this in turn increases the concentration of nitrogen as nitrate after pretreatment. All species of nitrifying bacteria require a number of micronutrients. Most important among these is the need for phosphorus for ATP (Adenosine Tri-Phosphate) production. The conversion of ATP provides energy for cellular functions. Phosphorus is normally available to cells in the form of phosphates (PO_4^{3-}). Nitrobacter, especially, is unable to oxidize nitrite to nitrate in the absence of phosphates. Although the maximum contaminant level for nitrogen as nitrate is 10mg/l, the average concentration of nitrogen as nitrate after pretreatment was 15mg/l which is higher than the maximum allowable limit. Thus, further treatment was needed to minimize the concentration of nitrogen as nitrate to bring towards the appropriate limit of discharge.

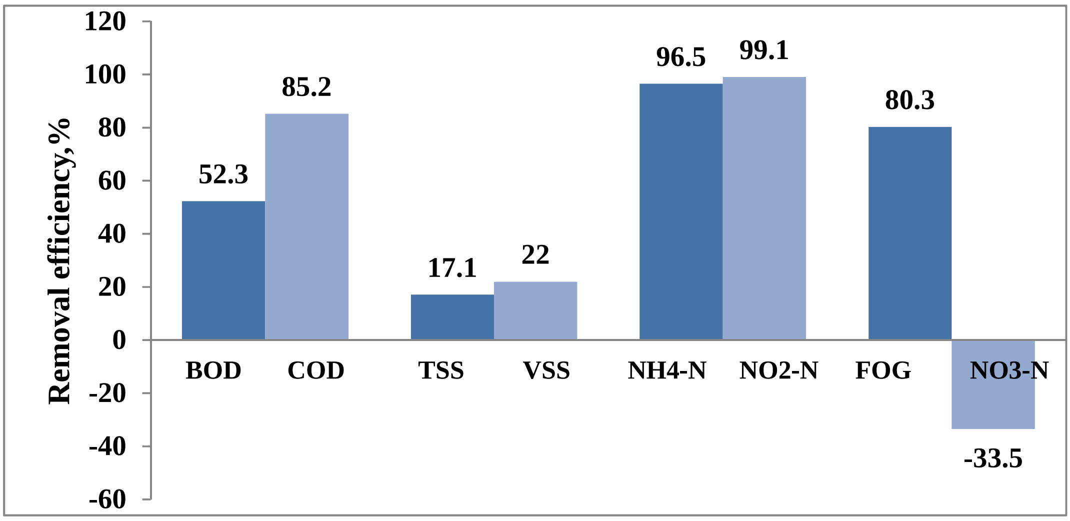

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Figure.4.3: Removal efficiency of sedimentation tank for the selected parameters

4.2. Characterization of Parameters for HSSFCWs

The dairy waste water characteristics and the removal performances of the three horizontal subsurface flow constructed wetlands were investigated, in which two of the wetlands were planted with the emergent plants *Typha Latifolia* and *Scirpus Lacustris*, the rest of the wetland was only gravel or unplanted. The successful application of constructed wetland technology to the treatment of wastewater with relatively low organic loadings has been well documented [1]. In this research project, a constructed wetland was utilized to study the treatment of dairy waste water that is highly organic and full of suspended solids. The objective of the wetland cells was to accomplish secondary treatment and this section will evaluate the performance of the wetland cells. After laboratory investigations of the different parameters it was found that the mean influent BOD_5 and COD concentrations were 241.4mg/l and 359mg/l, respectively. And also the mean daily organic loading rate values for BOD_5 and COD were 4.64 g/m^2.d and 6.90 g/m^2.d, respectively. The mean influent concentrations for nitrogen as TN, NO_3^-–N, NO_2^-–N, $NH4^+$-N were 39.4mg/l, 15mg/l, 0.002mg/l and 0.15mg/l, respectively. Accordingly, the mean organic loading rates for TN, NO_3^-–N, NO_2^-–N, NH_4^+-N were found to be 0.76 g/m^2.d,0.29 g/m^2.d,0.00004 g/m^2.d and 0.003 g/m^2.d respectively. Phosphorous as TP and PO_4^{3-}-P had mean influent concentrations of 7.8mg/l and 3.5mg/l, respectively.

In the same manner, the mean organic loading rates of TP and PO_4^{3-}-P were 0.15g/m^2.d and 0.07g/m^2.d, respectively. The mean influent concentrations of solids as TS, TSS and VSS were found to be 967mg/l, 265mg/l and 156mg/l, respectively. The organic loading rates of TS, TSS and VSS were 18.6 g/m^2.d,5.1 g/m^2.d and 3 g/m^2.d, respectively. And also the mean influent concentrations of FOG, TC and TCa were to be found 80mg/l, 835MPN and 64mg/l, respectively. The organic loading rates of these parameters were 2.09 g/m^2.d,16g/m^2.d and 1.23 g/m^2.d, respectively.

Table: 4.1. Raw, influent, effluent wastewater concentration and Organic loading rates

Parameter	Raw dairy wastewater (Mean ± SD)	Influent concentration (Mean ± SD)	Effluent concentration (Mean ± SD)			Organic loading rate ($g/m^2/d$)
			SSFC W_1 (TyphaLatifolia)	SSFC W_2 (ScirpusLacustris)	SSFC W_3 (control)	
BOD_5	506.2±70	241.4±32	47±19[a]	38±12[a]	84±26[a]	4.64
COD	2520.8±189	359±53	98±31[a]	56±18[a]	114±45[a]	6.90
TN	50.6±13	39.4±7	26.8±5[a]	22.3±4[a]	30.4±6[b]	0.75
NO_3^--N	0.58±0.15	15±6	0.31±0.05[a]	0.27±0.02[a]	1.7±0.4[b]	0.28
NO_2^--N	0.23±0.01	0.002±0.0001	0.05±0.002[a]	0.07±0.003[a]	0.04±0.0015[a]	0.000038
NH_4^+-N	4.35±2	0.152±0.009	0.035±0.00[a]	0.024±0.00[a]	0.062±0.004[a]	0.0029
TP	10.2±4.5	7.8±3.2	4.4±1.2[a]	3.6±1.2[a]	5.7±1.9[b]	0.15
PO_4^{3-}-P	5.3±1.8	3.64±0.8	0.8±0.02[a]	0.7±0.01[a]	0.9±0.03[b]	0.07
Ca	88±17	64±9	43±4[a]	39±4[a]	50±7[b]	1.23
EC,µs/cm	1187±83	987±72	764±53[a]	751±48[a]	773±62[a]	18.9
PH	5.7±0.3	7.4±0.5	8.4±0.7[a]	8.2±0.6[a]	8.6±0.8[a]	0.14
T,^{0}C	28±7	26±5	25±4[a]	23±1[a]	29±9[a]	-
DO	1.3±0.5	1.9 ± 0.1	2.4±0.2[a]	2.6±0.3[a]	2.1±0.2[b]	-
TC,MPN/ml	1062±73	835 ± 66	123±26[a]	86±11[a]	256±37[b]	16
FOG	406±29	80±15	4.9±0.2[a]	3.7±0.2[a]	5.6±0.4[a]	2.09
TS	1480±406	967±320	560±113[a]	510±96[a]	602±125[a]	18.6
TSS	318±20	265±17	85±10[a]	80±10[a]	93±12[a]	5.1
VSS	200±23	156±18	95±8[a]	89±5[a]	106±10[a]	3

Parameters with the same letter of **a** are not significantly different and when the letters are different the factors are significantly different on the respective parameters.

N.B: Except for T^0, pH, EC and TC all parameters are in mg/l

4.2.1. BOD and COD

Organic matter is decomposed in HSSFCWs by both aerobic and anaerobic microbial processes as well as by sedimentation and filtration of particulate organic matter. The average BOD_5 percentage removal efficiency of the two HSSFCWs planted with *Typha latifolia* and *Scirpus lacustris* was 80.5% and 84.2%, respectively. Accordingly, the average COD removal efficiency of *T.latifolia* and *S.lacustris* was found to be 72.7% and 84.4%, respectively.

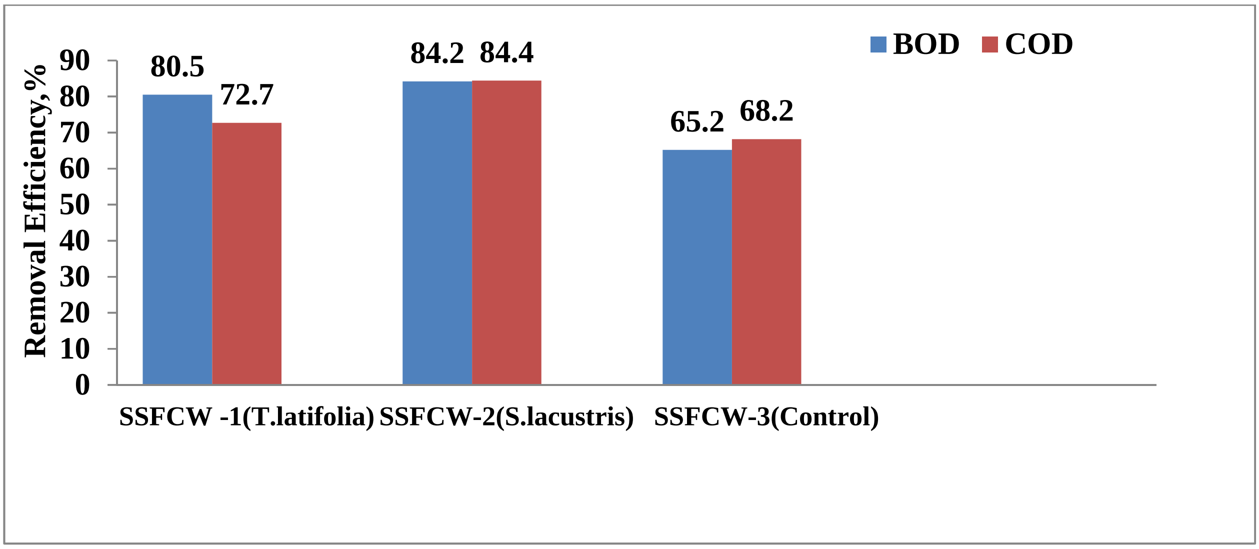

Figure.4.4: Removal efficiencies of the three HSSFCWs for BOD and COD

From these results the removal efficiency for BOD_5 and COD of the constructed wetland planted with *T.latifolia* was less than the wetland planted with *S.lacustris*. The reason for the decrease in the removal performance of the wetland planted with *T.latifolia* to the one planted with *S.lacustris* was that the roots of *S.lacustris* are highly diversified and many in number occupying larger area than *T.latifolia* plant species. The number of roots is very important for the survival of microorganisms that are important in the various conversion processes that need oxygen such as in degrading organic matter and oxidation by providing air movement space. As a result of the organic matter degradation using the wetland planted with *S.lacustris* the concentration of BOD_5 and COD decreased relatively which increased the removal efficiency of the plant. Even though there was difference in the removal efficiency of the two plant species, there was not any statistical significant difference

(P>0.05) observed between their removal performances for BOD_5 and COD. The control unit of the wetland relatively showed lower removal efficiency compared to the planted wetlands that is 65.2% and 68.2% for the parameters BOD_5 and COD, respectively. These values indicated that the difference in removal efficiency between planted and unplanted constructed wetlands was found not statistically significant (P> 0.05).Generally it was observed that the dissolved oxygen (DO) concentration increased after the dairy waste water passed the three HSSFCWs. After the influent waste water passed the wetland planted with *S.lacustris* the DO concentration increased by 26.9% and also the wetland planted with *T.latifolia* increased the DO concentration in the wastewater by 20.8%. Unike the planted wetlands DO concentration showed a small increase of 9.5% after the waste water passed through the unplanted wetland. The ANOVA and LSD tests indicated that there is significant difference (P<0.05) between the planted and unplanted HSSFCWs for DO.

4.2.2. TSS and VSS

From Table 4.1 it was observed that the average TSS removal efficiency of constructed wetland planted with *T.latifolia* was found to be 68% and that of planted with *S.lacustris* was 70%.Accordingly, the average removal efficiency of wetland with *S.lacustris* and *T.latifolia* for VSS was 43% and 39%, respectively.

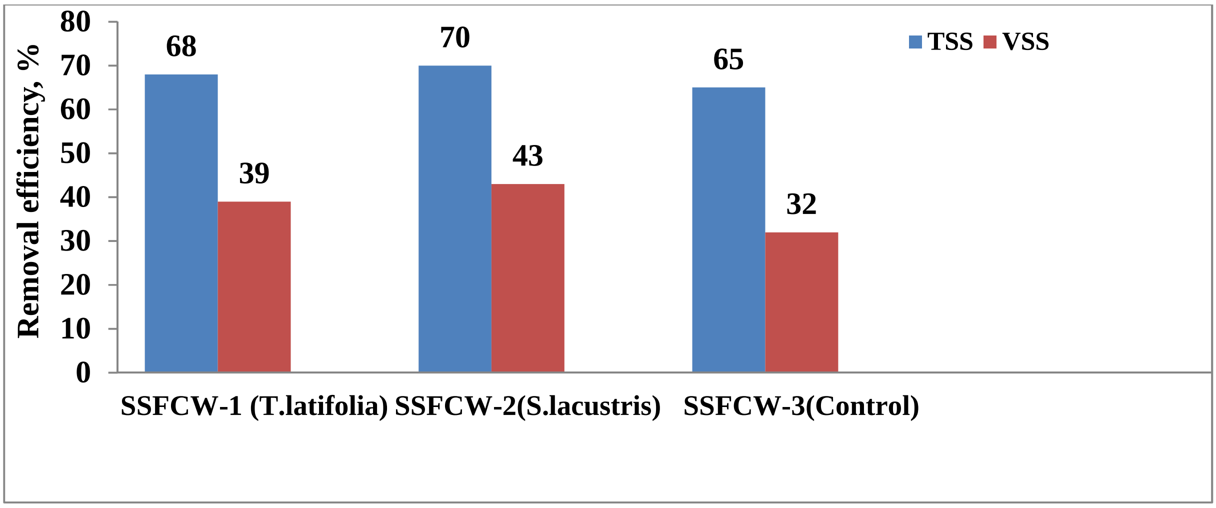

Figure.4.5: Removal efficiencies of the three HSSFCWs for TSS and VSS

These values indicated that the removal performance of constructed wetland vegetated with *S.lacustris* had higher performance than the one planted with *T.latifolia*. As it was mentioned the roots of the plant species *S.lacustris* are more diversified than that of *T.latifolia* to block the movement of the suspended solids that minimizes the effluent concentration and hence increasing its removal efficiency. It was found that the two planted wetlands had no significant difference ($P > 0.05$) in the removal of TSS and VSS. The wetland used as a control showed close removal performance to the two wetland plants that is 65% and 32% for TSS and VSS, respectively. The difference in the removal performance of the wetland planted to the unplanted showed that there was not statistical significant difference ($P > 0.05$) for TSS and VSS.

4.2.3. TN, NH_4^+-N and NO_3^--N

Nitrogen as TN, NH_4^+-N and NO_3^--N showed significant decrease in concentration after the wastewater was treated by the two vegetated constructed wetlands. The average removal efficiency of the wetland planted with *S.lacustris* showed relatively higher removal performance of 43.4%, 84.2% and 98.2% for TN, NH_4^+-N and NO_3^--N, respectively than the wetland planted with *T.latifolia*, that is, 32%, 77%and 97.5% for the parameters TN, NH_4^+-N and NO_3^--N, respectively.

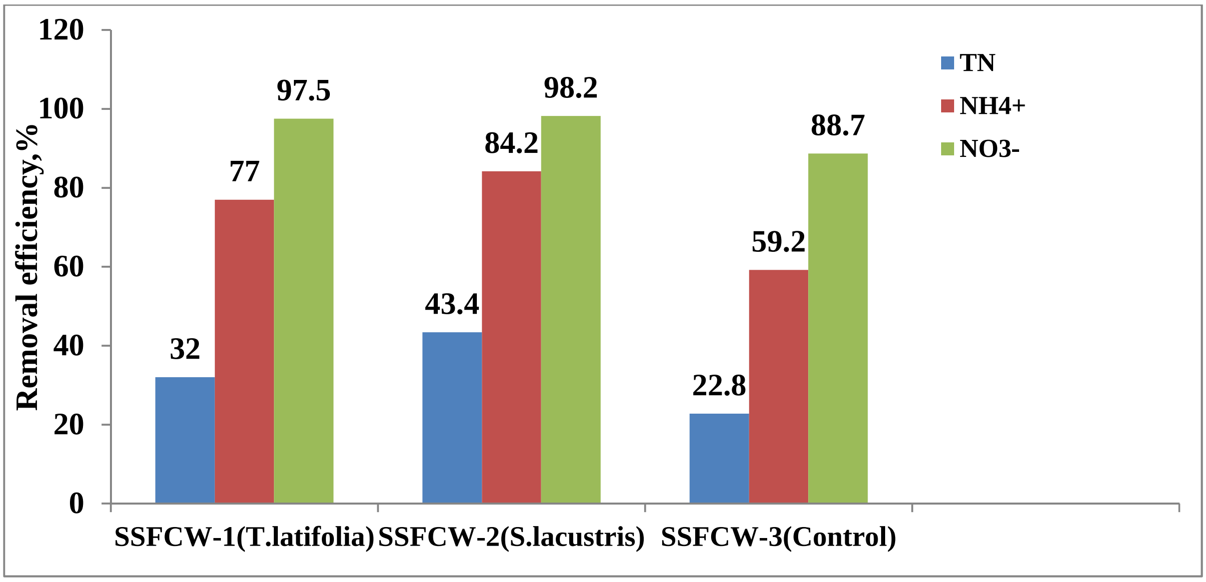

Figure.4.6: Removal efficiencies of the three HSSFCWs for TN,NH_4^+-N and NO_3^-

The most important conversion processes functioning in a wetland system are: ammonification (organic N → NH_4^+), nitrification (NH_4^+→ NO_2^-→NO_3^-), denitrification (NO_3^- → N_2O → N_2), biological fixation (N_2→organic N), nitrate ammonification (NO_3^- → NH_4^+), anaerobic ammonia oxidation (ANAMMOX, NH_4^+→N_2) and volatilization (NH_4^+→ NH3). The microbial processes involved in nitrogen transformations have traditionally been described by aerobic nitrification, anoxic/anaerobic denitrification and aerobic/anaerobic ammonification. However, intensive studies, often aimed at processes responsible for nitrogen removal from wastewaters revealed many other microbial processes responsible for nitrogen transformations such as anaerobic ammonia oxidation, aerobic denitrification [1].The reason for the decrease in the removal performance of the wetland planted with *T.latifolia* than the one planted with *S.lacustris* was that the number of *S.lacustris* plant species planted for the experiment were more in numbers than *T.latifolia* for *T.latifolia* roots grow horizontally and possess rhizomes that are important for reproduction. This plant species need more space while planting hence decreasing the number of *T.latifolia* plant species in the wetland. Unlike *T.latifolia* the number of roots is more in *S.lacustris* and they also penetrate more depth than *T.latifolia*. Thus due to the existence of more number of microorganisms in the roots of *S.lacustris* important for the conversion of Nitrogen or assimilation of Nitrogen for protein synthesis. The decrease in the concentration of NH_4^+ in the wetlands was due to volatilization and anaerobic ammonia oxidation, that is direct conversion into ammonia gas and into nitrogen gas, respectively. Nutrient removal in systems with free-floating plants is far more complicated than plant uptake alone. Nitrogen is removed through plant uptake (with regular harvesting), ammonia volatilization, and nitrification-denitrification. The nitrifiers can flourish attached to the roots, which provide oxygen [6]. Nitrification also occurs in the water column when dissolved oxygen levels of the water are adequate to support activity of nitrifying bacteria. These conditions are usually created at relatively low plant densities and a partial plant cover over the water surface. Denitrification is the most significant and common process for NO_3^- removal in wetlands. Compared to the removal performance of the planted wetlands the control unit of the wetland showed less performance in removal of pollutants. The removal efficiency of the control unit of the wetland for TN, NH_4^+-N and NO_3^--N were 22.8%, 59.2% and 88.7%, respectively. When carrying out ANOVA and LSD tests for these differences in performance the planted and the control SSFCWs for TN and NO_3^--N were found significantly different ($P<0.05$) and for NH_4^+-N their removal performance difference were not significant ($P>0.05$).From Table 4.1.it was seen that the uptake of TN, NH_4^+-N and NO_3^--N by *S.lacustris* were 12.1mg/l,0.038 mg/l and 1.43 mg/l, respectively. And also the uptake of TN, NH_4^+-N and NO_3^--N by *T.latifolia* were 3.6 mg/l, 0.027 mg/l and 1.39 mg/l, respectively.

4.2.4. TP and PO_4^{3-}-P

From Table.4.1, the removal performance of the wetland planted with *S.lacustris* for TP and PO_4^{3-}-P was53.8% and 80.7%, respectively. And also TP and PO_4^{3-}-P were removed from the wetland planted with *T.latifolia* with the efficiency of 43.5%and 78%, respectively. And the removal performance of the wetland planted with *S.lacustris* was relatively higher than the wetland planted with *T.latifolia* this is because *S.lacustris* plant species roots can relatively penetrate greater depth and they are more in numbers to absorb phosphorous as PO_4^{3-}-P. And also the control unit of the wetland had removal efficiency of 27% and 75.2% for the parameters TP and PO_4^{3-}-P, respectively. From Table 4.1 the uptake of TP and PO_4^{3-}-P for the plant species *S.lacustris* were found 2.2 mg/land 0.2 mg/l, respectively. And also the uptake of TP and PO_4^{3-}-P for *T.latifolia* were found to be 1.3 mg/l and 0.1 mg/l, respectively.

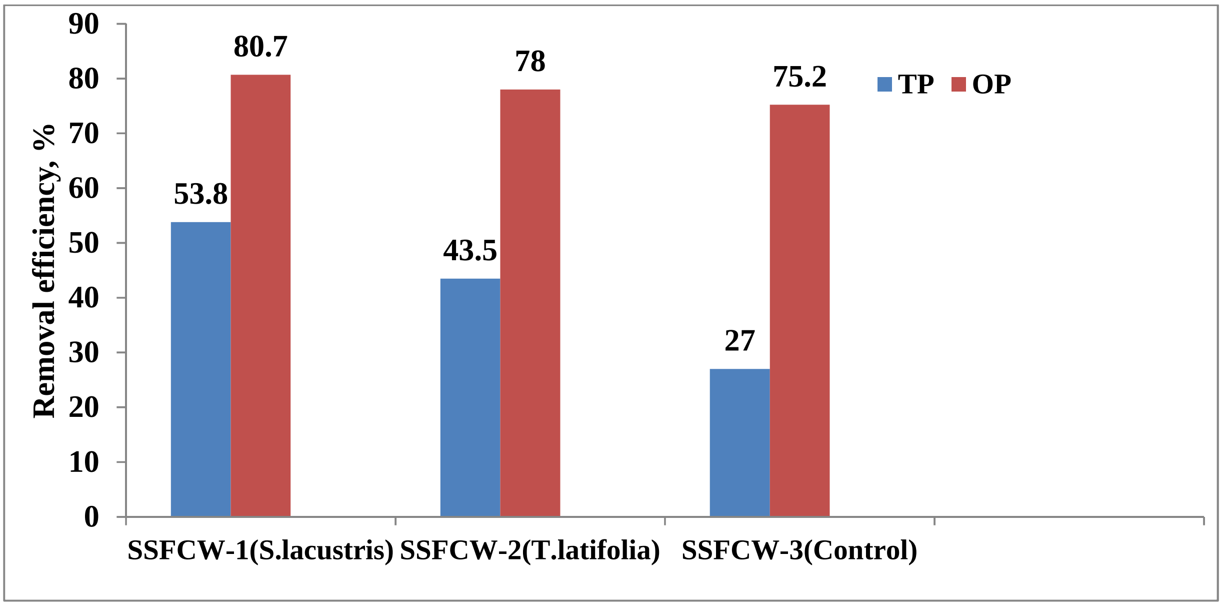

Figure 4.7: Removal efficiencies of the three HSSFCWs for TP and OP

Phosphorous is an important plant macronutrient, making up about 0.2% of a plant's dry weight. It is a component of key molecules such as nucleic acids, phospholipids, and ATP, and, consequently, plants cannot grow without a reliable supply of this nutrient. Inorganic Phosphorous is also involved in controlling key enzyme reactions and in the regulation of metabolic pathways. After N, P is the second most frequently limiting macronutrient for plant growth. Although the total amount of Phosphorous in the soil may be high, it is often present in unavailable forms. In the soil more than 80% of the Phosphorous becomes immobile and unavailable for plant uptake because of adsorption, precipitation, or conversion to the organic form [59]. Phosphorus can be removed from these systems by microbial assimilation, precipitation with divalent and trivalent cations, or adsorption onto clays or organic matter. However, most studies have shown that plant uptake and subsequent harvest is the only reliable long-term Phosphorous removal mechanism. Harvesting is essential because detritus of plants tend to release Phosphorous into water during decomposition, thus, decreasing the Phosphorous removal efficiency of the system [59].

4.2.5. Fat, Oil and Grease, Total Calcium and Total Coliform

From Table 4.1, it was observed that the removal efficiency of the wetland planted with *S.lacustris* for the parameter FOG was 95.3%. And the wetland planted with *T.latifolia* showed removal efficiency of 93.8%. In the same manner the control unit of the wetland had removal efficiency of 93% for the parameter FOG. Fat, Oil and Grease have the possibility to attach on the roots of the plants and on the surface of the gravel. After carrying out ANOVA and LSD tests it was found that there was not any significant difference ($P>0.05$) in the removal of FOG between the planted and unplanted HSSFCWs. Dairy factory wastewater commonly contain milk, byproducts of processing operations, cleaning products and various additives that may be used in the factory. Milk typically contains water (87%), fat (4%), protein (3.5%), lactose (4.7%) and ash (0.8%) [7].The fat content ranges from 3 - 5% with the major component being triglycerides (98%). Other fat components include phospholipids (0.5 - 1% of fat) and sterols (0.2 - 0.5% of fat) [28].

The HSSFCW planted with *S.lacustris* showed removal efficiency of 39% for total calcium (TCa). And also the wetland planted with *T.latifolia* removed the total calcium with efficiency of 32.8% which is better than the unplanted wetland with removal efficiency of 21.8% for total calcium. From Table 4.1 it was observed that the uptake of TCa by *S.lacustris* was found to be 11 mg/l and also the uptake of TCa by *T.latifolia* was found to be 7 mg/l. The role of calcium in plants is quite similar to that in people; it is, essential for good growth and structure. Insufficient calcium levels lead to deterioration of the cell membrane; the cells become leaky resulting in the loss of cell compounds and eventually death of the cell and plant tissue. Calcium, in addition to its role in cell structure, also plays a role in regulating various cell and plant functions as a secondary messenger. This function as a secondary messenger assists in various plant functions from nutrient uptake to changes in cell status to help the plant react to the impact of environmental and disease stresses. Table 4.1 showed also that the three HSSFCWs were good at the removal of Total Coliform (TC) during their HRT of 8 days, that is, the HSSFCW planted with *S.lacustris* removed the TC with the efficiency of 89.7% and the one planted with *T.latifolia* had removal performance of 85.2%.The unplanted HSSFCW removed the TC in relatively lower capacity of 69.3%%. From these values the ANOVA and LSD tests were performed and it was found that there was significant difference($P<0.05$) between the planted and unplanted HSSFCWs for TC. Wastewaters contain various pathogenic or potentially pathogenic microorganisms which, depending on species concentration, pose a potential risk to human health and whose presence must therefore be reduced in the course of wastewater treatment [78]. It was suggested that HF constructed wetlands are able to remove both bacteria and viruses from sewage with exponential decay with respect to distance along the channel [79]. A clear dependence of FC removal on HRT was reported and it was observed that the increase of FC removal for HRT of 2 and 9.5 days, respectively. Root systems also release other substances besides oxygen. These substances are usually organic compounds such as anaerobic metabolites, organic acids, phytometallophores, peptides (e.g.phytochelatins), alkaloids, phenolics, terpenoids or steroids. The magnitude of this release is still unclear, but reported values are generally in the range of 5-25% of the photo synthetically-fixed carbon. Functions of the root exudates are manifold. Allelopathy, i.e., inhibition of one plant species through chemical means by another plant has been well documented in wetlands. Allelopathic chemicals belong to several chemical groups, particularly phenolics, organic acids, plant hormones and plant metabolites of plant-aromatic amino acids, alkaloids, flavonids, terpenoids, steroids, long-chain fatty acids, and elemental sulfur [79].Their importance for the process of wastewater treatment is the release of anti-microbial compounds by roots of wetland plants. One of the first studies reporting on the excretion of anti-bacterial substances by macrophytes was activity by alkaloids extracted from *Nupharlutea*. It was showed that the *Scirpuslacustris* (Bulrush) releases antibiotics from its roots and range of bacteria (coliforms, salmonella and enterococci) obviously disappeared from polluted water by passing through a vegetation of bulrushes. No perfect indicators have been found, but the coliform bacteria group has been long used as the first choice among indicator organisms. Coliforms are usually monitored as total or fecal coliforms. Total coliforms (TC), however, are ubiquitous in surface waters, and they include many bacteria from the family *Enerobacteriaceae* that are not derived from human or other animal pollution sources.

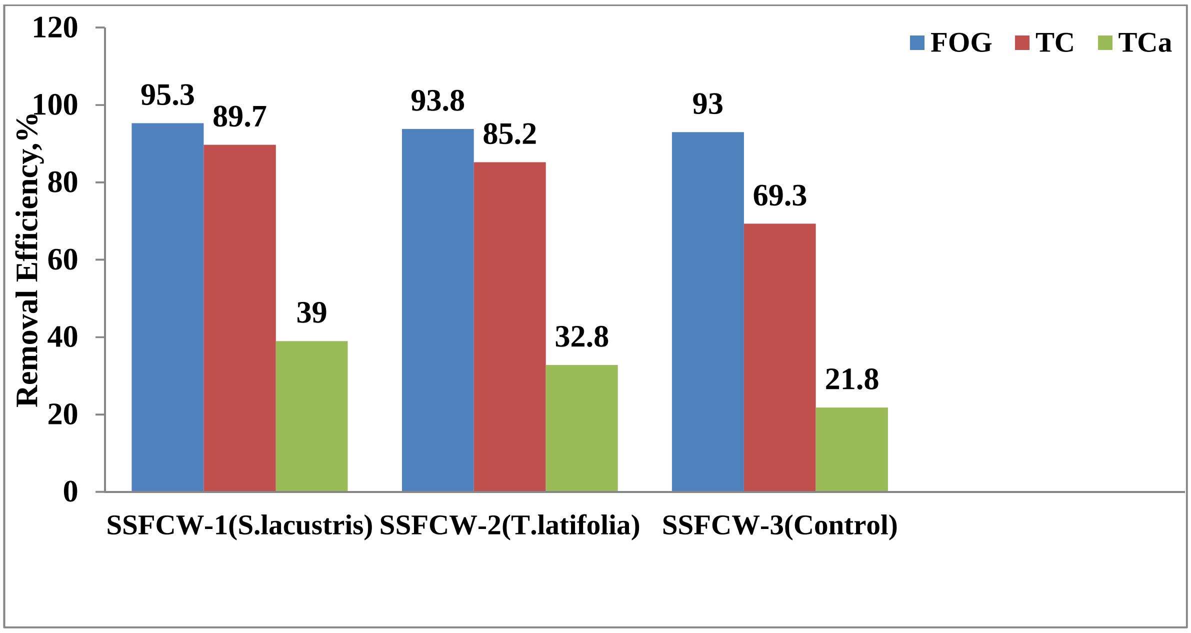

Figure.4.8: Removal efficiencies of the three HSSFCWs for FOG, TCa and TC

Chapter 5

Conclusion and Recommendations

5.1. Conclusion

One of the primary objectives of this research was to determine the treatment capacity of dairy wastewater by HSSFCWs planted with *Typha Latifolia* and *Scirpus Lacustris*. The primary treatment unit basically is helpful in removing solids by gravitational settling process. The pretreatment unit attained a performance of 52.3% and 85.7% for BOD and COD and 17.1% and 22% for TSS and VSS before discharging to the constructed wetland cells. From these results the pretreatment unit performed less in removing the Organics so that the BOD and COD values didn't comply with the standards but it performed well in removing the solids; TSS and VSS values complied with the standards. The wetland cells removed the pollutants effectively than the pretreatment unit to the extent that the parameters comply with the standards. The HSSFCW planted with *Scirpus Lacustris* relatively showed good removal capacity than the rest two CWs.

Accordingly, the HSSFCW planted with *Scirpus Lacustris* showed better removal capacity than HSSFCW planted with *Typha Latifolia* for the roots of the former are many in number and occupy more space than the latter that is suitable for microorganisms' growth. The HSSFCW that performed better in removing the pollutants got average removal efficiencies 84.2%, 84.4%, 43% and 70% for BOD, COD, TSS and VSS, respectively. And for TN, NH_4^+-N, NO_3^--N, TP and PO_4^{3-}-P the efficiencies were 43.4%, 84.2%, 98.2%, 53.8% and 80.7%, respectively. The FOG, TCa and TC got their higher efficiencies of 95.3%, 39% and 89.7%, respectively. The values of pH, Temperature, DO and EC after the influent passed through the wetland planted with *Scirpus Lacustris* showed higher average removal efficiencies. From these values it can be deduced that the HSSFCW planted with *Scirpus Lacustris* showed higher removal performance than the one planted with *Typha Latifolia*. In addition to their wastewater treatment capacity *Scirpus Lacustris* and *Typha Latifolia* plant species have medicinal and dietary values, thus the versatile advantages of these plant species is promising that makes us give due attention to protect our vulnerable environment and human health from the threat a dairy wastewater discharge could pose and their dietary values to solve our food insecurity to some extent. From this research it was observed that the main factors for the pollution of dairy wastewater were BOD, nitrogen (as TN, NH_4^+-N and NO_3^--N) and phosphorous (as TP and OP). Some part of Horra Lake near Ada milk factory is covered with algae posing fatality for fishes and unfavorable condition for some important aerobic microbes by blocking the sunlight from penetrating the water body and minimizing the DO by shielding the interface of water-air contact. Thus, it is evident and mandatory to apply environmentally friendly and financially feasible treatment system like constructed wetlands.

5.2. Recommendations

Due to the high loading rates of dairy wastewater, much damage to the system performance can be done due to a non-functioning settling basin. It would be recommended that the settling basin consist of two or more continuous deep basins in parallel. These basins should also include scum baffles in order to retain the lighter solids, fats and scum from leaving the basins. If these basins were sufficiently sized, the time and cost of maintaining the basins could be decreased. Another alternative would be to have solid pump or other available appropriate solids removal device so that solids could be removed on a more frequent basis. The cost and maintenance of the solids removal device would be, weighted against the nutrient reduction benefits received by removing the complexed, nutrients before they become dissolved. Once removed the solids need to be properly managed. Best management practices would include land spreading or, composting. Recommended changes to the wetland portion of the system would be as follows: - To enhance the removal of nutrients within the wetland cells could be separated by stretches of shallow, open water areas. These areas would allow greater reaeration of the water, which would be more conducive to nitrification, followed by denitrification in the more anaerobic wetland cells downstream.

The number and placement of these shallow open waters has not been studied. Another option to investigate would include the recirculation of the wastewaters back to the anaerobic wetland cells. This option would attain some of the same goals of providing anaerobic and aerobic conditions; however the greatest setback would be the associated costs of purchasing, maintaining and operating this type of system. Recommendations for future studies can also be provided. The primary one is the need to understand the fate and transport of nitrogen through a constructed wetland treating dairy wastewater. One of the additional water parameters necessary to accomplish this would be testing for NBOD along with BOD. Another difference may be attributed to the presence of greater concentrations of ammonia, which in the presence of nitrifying bacteria produce a nitrogenous biological oxygen demand (NBOD). The NBOD may produce lower BOD_5 results, if sufficient numbers of nitrifying bacteria are present. NBOD testing was not performed during this study, so this possibility could not be confirmed. Due to the apparent high levels of nitrogenous compounds in the dairy wastes it can be suggested that future similar studies perform NBOD testing concurrent with BOD_5 tests. Attaining this additional information would give the investigator an indication of the system organisms' presence and ability to accomplish nitrogen reductions. Given this information one may get some indication of the true advantage of including an alternating anaerobic and aerobic sequence to the wetland cells. The last but not the least thing to recommend is due to climatic and natural factors, the emergent plants chosen for the dairy wastewater treatment could not achieve acclimatization stage with in the short research period resulting in relatively weak performance.

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Appendices

Appendix-I: Collection of *Scirpus lacutris* and *Typha Latifolia* plants from Ziway Lake

Appendix-II: Laboratory measuring procedures for TN

Persulfate measuring method for TN

1. Add one total nitrogen persulfate reagent powder pillow to each of two total nitrogen hydro

Oxide reagent

2. Add 2ml of sample to one vial and add 2ml of distilled water (to another vial-the blank)

3. Cap both vials and shake vigorously.

4. Place the vial in the COD reactor and heat for 30 minutes.

5. Enter the stored program number for test N tube TN.

6. Rotate the wavelength dial until the small display shows when the correct wavelength is dialed in the display will quickly show zero sample.

7. Remove the caps from the cooled digested vials and add the contents of one TN reagent a powder pillow to each vial .Cap vials and shake for 15 seconds.

8. After the beeps, remove the caps from the cooled vials and add on TN reagent B powder pillow to each vial. Cap vials and shake for 15 seconds.

Reagents	Quantity required per test	Unit
TN hydroxide reagent, 0.1 N	2 vials	25/Kg
TN persulfate reagent	2 pillows	100/Kg
TN reagent A powder pillows	2 pillows	100/Kg
TN reagent B powder pillows	2 pillows	100/Kg
TN reagent C vials	2 vials	25/Kg

Appendix-III: Laboratory measuring procedures for TP

Acid persulfate digestion method for TP

1. Measure 25ml of sample into a 50 ml (or larger) Erlenmeyer flask using a graduated cylinder.

2. Add the contents of one poatassium persulfate powder pillow and swirl to mix.

3. Add 2ml of 5.25N Sulfuric acid solution.

4. Place the flask on a hot plate and boil gently for 30 minutes.

5. Cool the sample to room temperature.

6. Add 2ml of 5N sodium hydroxide solution and swirl to mix.

7. Pour the sample into a 25 ml graduated cylinder

8. Proceed with a reactive phosphorous method that will cover the expected TP concentration range.

Reagents	Quantity required per test	Unit
Potassium persulfate powder pillows	1 pillow	100/Kg
Sodium hydroxide solution, 5 N	2ml	100ml
Sulfuric acid solution, 5.25N	2ml	100ml