International Journal of Environmental & Science Education, 2016, **11**(6), 767-777

Determination of the Fate of Dissolved Organic Nitrogen in the Three Wastewater Treatment Plants, Jordan

Mohammed Wedyan & Ahmed Al Harahsheh & Esam Qnaisb
Biological Sciences and Biotechnology Department, The Hashemite University, JORDAN

•*Received 28 January 2016* •*Revised 29 March 2016* •*Accepted 30 March 2016*

This research aimed to assess the composition of total dissolved nitrogen (TDN) species, particularly dissolved organic nitrogen (DON), over the traditional wastewater treatment operations in three biological nutrient removal (BNR) wastewater treatment plants (WWTPs) in Jordan. It had been found that the DON percentage was up to 30% of TDN within the effluent plant;which restricted the plant's ability to eliminate nitrogen to minimal amounts. Effluent DON levels from the three plants varied from 11.7 to 34.8 mg N/L and would not fluctuate substantially, even if there seemed to be a substantial difference inside influent organic nitrogen levels. The main transforming of DON and biodegradable dissolved organic nitrogen (BDON) along the treatment train had been noticed in the aerobic method. More than 70% of effluent DON was consisting of hydrophilic materials, which promote algal growth. The research presented significant information for foreseeable future improving of WWTPs or the choice of DON elimination techniques to satisfy additional challenging nitrogen release limits.

Keywords: total dissoloved nitrogen, dissolved organic nitrogen (DON), bioavailable/biodegradable dissolved organic nitrogen, wastewater treatment plants, Jordan.

INTRODUCTION

The wastewater effluent can be an important source of anthropogenic nitrogen to aquatic environments and possesses undesirable influences over the water quality, particularly in effluent-dominated waters (Pagillaet al 2008, Bronk et al 2010, Liu et al 2012).In order to minimize the eutrophication with the aquatic environment, biological enhanced nitrogen removal (BENR) operations that eliminate the majority of the dissolved inorganic nitrogen tend to be regularly used within wastewater treatment (Czerwionka et al 2012). Because of successful of BENR processing, the most important part of the remaining nitrogen within effluent released to the responsive environments is made of dissolved organic nitrogen (DON) (Pagilla et al 2006, Liu et al 2012). Earlier reports revealed that effluent DON is usually

Correspondence: Mohammed Wedyan,
Biological Sciences and Biotechnology Department, The Hashemite University, Al Zarka, P O Box 330127, Jordan
E-mail: mwedyan@hu.edu.jo
doi: 10.12973/ijese.2016.344a

ISSN: 1306-3065

bioavailable to algae and also plankton (Pehlivanoglu-Mantas and Sedlak2004, Sattayatewa et al 2009, Simsek et al 2013). Accordingly, DON is actually drawing a greater number of interesting in waste water treatment plants (WWTPs) recently as a result of increasing worries including the revitalizing algal continuing growth of aquatic environments (Pehlivanoglu-Mantas and Sedlak 2004) and also the forming nitrogenous disinfection by-product N-nitrosodimethylamine (Pehlivanoglu-Mantas and Sedlak 2006, 2008, Lee et al2007). The rising issues regarding wastewater-derived DONhave raised the necessity to know more about their levels, compositions and properties.

DON is usually driven by subtracting dissolved inorganic nitrogen (DIN, the sum of ammonium, nitrate and nitrite) concentrations out of the total dissolved nitrogen(TDN) concentrations. Low DON concentration within waters abundant with proportion of DIN to TDN using the available techniques is commonly imprecise and DON estimating frequently possess high standard deviations (Lee and Westerhoff 2005, Vandenbruwane et al 2007). In order to improve the precision and accuracy of DON estimating, many pretreatment techniques were applied to eliminate DIN species in waters, for example dialysis (Lee and Westerhoff 2005) and nano filtration (NF) (Xu et al 2010). The restricted accessible techniques frustrated researchers looking into the characteristics and behavior of DON in wastewater treatment plants. As a substitute for determining DON as being a big issue in wastewater, research study has quantified certain organic nitrogen made up of substances like dissolvedfree and combined amino acids (DFAA and DCAA), dimethyl amine (DMA), and ethylenediaminetetraacetic acid (EDTA) (Pehlivanoglu-Mantas and Sedlak 2008).

Previous reports demonstrated that about 70% of waste water-derived DONalso cannot be described with available techniques (Pehlivanoglu-Mantas and Sedlak 2006, 2008, Simsek et al2012). Earlier investigations on wastewater-derived DON focused on two questionable problems; where DON is eliminated or even created in BENR processes and what exactly is the influence of BENR process on DON (Czerwionka et al 2012,Simsek et al 2013, Huo, et al., 2013). Sattayatewa et al (2009) reported that around 28-57% of the effluent DON was bioavailable or biodegradable, Simsek and his colleague (2013) determined biodegradable dissolved organic nitrogen (BDON) and bioavailable dissolved organic nitrogen(ABDON) in activated sludge (AS) and trickling filter (TF)wastewater treatment processes. They recorded that 65%BDON and 63% ABDON had been eliminated in the TF facility and 68%, 56%, respectively within the AS facility. Understanding the behavior of DON across the biological processes inside the WWTPs plays important role in realizing the function of treatment units in the particular elimination of DON. Even so, present data around the qualities of DON in BNR WWTPs continue to be constrained and inadequate to know the future or alterations of DON over the activated sludge process (Czerwionka et al 2012).

In Jordan many farmers are turned to treated wastewater as a low-cost alternative to conventional irrigation freshwater. However, wastewater is rich in essential nutrients such as nitrate, nitrite, ammonia, organic nitrogen and phosphate which lowers the cost of fertilizers additives. Meanwhile, wastewater used for irrigation may possess a health risk to peoples as it may contain excessive nutrients. Therefore, this study comes to highlight the risk of high organic nitrogen contents in three main waste water treatment plants that were investigated; Al Zarqa, Mafraq, and Irbid to achieve more information on wastewater-derived DON in WWTPs in Jordan. And to assess the effect of biological treatment on DON and its components. Finally, provide a better understanding of the fate of the unidentified DON WWTPs.

MATERIALS AND METHODS

Sample sources

Samples were obtained from three different treatment plants effluent, which are Al-Zarqa (ZTP), Irbid (ITP), and al Mafraq (MTP) (Figure 1). The selected plants treat about 90% of the domestic wastewater in Jordan and serve more than 2 million inhabitants (Bataineh et al., 2002), the treatment process shown in Table 1.

All plants have to comply with the discharge limits for biochemical oxygen demand (BOD) and ammonia (based on the receiving river flow rate) but are not subject to any total nitrogen or total phosphorus limits.

Sample collection and preparation

The samples were collected from all of the WWTPs during different seasons, specifically February, April , and June. All samples were collected in polyethylene containers (acid-washed and rinsed with ultrapure water) (Mill-Q, Millipore Corp. USA) before used, then were delivered to the laboratory on ice, filtered through 0.45 μm cellulose acetate membranes upon arrival, and then were stored at 4°C in the dark.

Dissolved inorganic Nitrogen (DIN) species

Total dissolved nitrogen was measured with the standard persulfate digestion method (APHA, 1998). Nitrate concentrations were measured with a ion chromatograph (equipped with Ion Pac AS-14 4x250mm column), after conversion of all the nitrogen forms to nitrate instead of the standard Cd-column reduction method (APHA, 1998). Nitrate and nitrite in undigested samples also were measured with the aforementioned ion chromatographic method. Ammonium (NH_4^+) was measured with the standard phenate method (APHA, 1998).

Dissolved Organic Nitrogen (DON) Calculation and Biodegradable Dissolved Organic Nitrogen (BDON) determination procedures

DON was calculated as the difference between the total dissolved nitrogen and the sum of inorganic nitrogen species (i.e., NO_3^+, NO_2^+, NH_4^+) using equation (1). The BDON procedure is as follows. All the samples were filtered through a 0.2 mm pore-size cellulose acetate membrane filter (WhatmanInc) within an hour after collection. A portion of the filtered samples were used for immediate analysis of total nitrogen and inorganic nitrogen species (ammonia, nitrite, and nitrate). The value was recorded as initial DON (DONi). Two hundred milliliters of the remaining filtered sample were mixed with 2 mL of acclimated inoculum bacteria in a 250 mL amber bottle. Raw wastewaters (collected from locations) were used as the inoculum.

Table 1. The treatment plants concerned in the study (Bataineh et al., 2002).

Plant	Treatment process	Remarks
Al-Zarqa	Stabilization ponds (natural aeration, facultative, anaerobic lagoons)	
Irbid	Screen, grit removal, primary sedimentation, biological process, secondary sedimentation, disinfections	Trickling filter & activated sludge
Mafraq	Screen, grit removal, biological process, secondary sedimentation, Polishing pond, infiltration, disinfections	Activated sludge with nitrogen removal technique

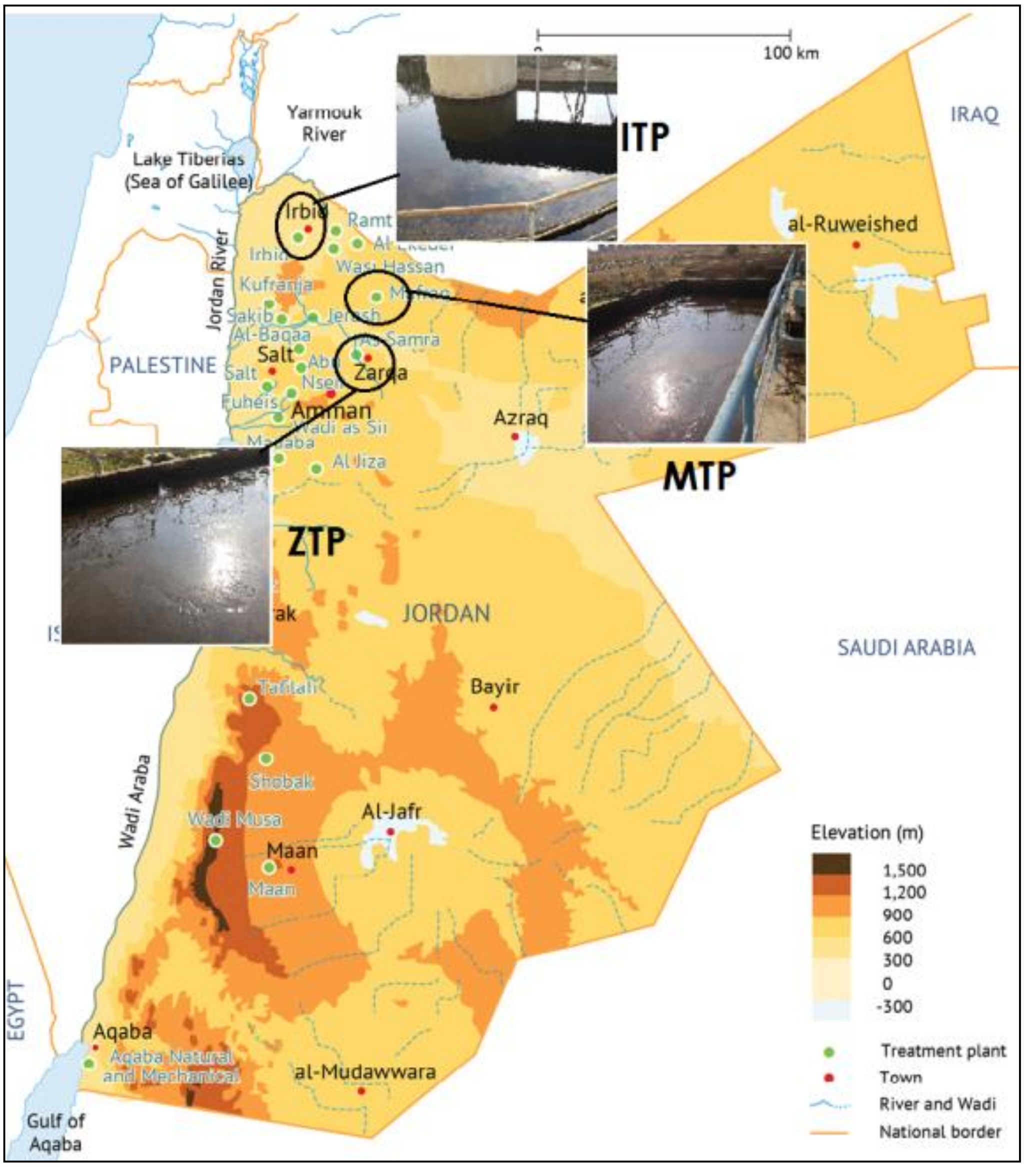

Figure 1. Jordan map indicating the sampling site

The solution in the bottle was shaken thoroughly to aerate and placed in an incubator in the dark at 20 °C for 16 days. During the incubation period, the solution in the bottle was manually shaken to aerate at least once every day to maintain aerobic conditions. A seed control (sample b), which was treated the same way as the samples, were prepared by adding the inoculum to 200 mL of de-ionized distilled water. After 16 days of incubation, all nitrogen species in the supernatant were measured to determine final DON (DON_f). BDON was calculated according to equation (2).

$$DON\ (mg/L\ as\ N) = TDN - DNH_3 - DNO_2 - DNO_3 \quad (1)$$
$$DON\ (mg/L\ as\ N) = (DON_i - DON_f) - (DON_{bi} - DON_{bf}) \quad (2)$$

where DNH_3, DNO_2, and DNO_3 are dissolved ammonia, nitrite and nitrate, respectively; DON_i and DON_f are DON before and after incubation for samples; and DON_{bi} and DON_{bf} are DON before and after incubation for control.The rate constant for organic nitrogen degradation was evaluated and computed by measured rate of disappearance of the DON using the equation: $dN/dT=-Kn\ .N_o$(Avnimelech et al 1995).

RESULTS AND DISCUSSIONS

The profile of different dissolved nitrogen species (ammonia, nitrite, nitrate, total nitrogen and organic nitrogen) in the three different wastewater treatment plants across the northern part of Jordan (Table 2).

Due to the environmental and weather conditions the concentrations of the nitrogen were fluctuation.

Dissolved nitrogen species

Dissolved total nitrogen (DTN)

The results show that the average concentration was ranged from 19.03 to 41.80 (mg - N/L). The highest concentration of DTN was found in ITP sample with average was 41.8±3.39 (mg/L ± SD) and the lowest concentration was detected in ZTP with average 19.30±6.60 (mg/L ± SD) (Table 2).

Dissolved Inorganic Nitrogen (DIN) species

All the nitrogen species in the dissolved fraction from effluent were detected the collected samples during this study (Table 2). The data show that the average concentrations of NH_4-N 0.039 ± 0.003, 0.06 ±0.019 and 0.70 ± 0.048 (mg-N/L ± SD) (ZTP, MTP and ITP, respectively). A small variation in ammonia concentrations (3%) was typically observed in the samples collected from ZTP and MTP. But highly significant difference with ITP samples was (85%). The plants process achieved almost complete ammonia removal during the wastewater treatment processes in three different plants, process in the ITP does not remove ammonia may be because of the toxicity of high oxygen concentration to the nitrifying microorganisms (Uemoto et al., 2000). Similar to the Fargo WWTF results, ammonia inall the samples were totally nitrified during the incubation (Pehlivanoglu-Mantas and Sedlak, 2008; Simsek et al, 2012, Liu, et al., 2013).

The results show that the highest concentration of NO3 was in MTP sample with average was 7.83±6.49 (mg/L ± SD) and the lowest concentration was detected in ITP with average was 6.33±3.80(mg/L ± SD) (Table 2).Nitrite was present at very low concentration in all WWTP. Generally, nitrite concentration was always less than 0.8 mg N/L in all collected samples. The highest concentration of NO2 was in ITP samples with average 0.7±0.48 and the lowest concentration was detected in MTP 0.013±0.002 (mg/L ± SD).Nitrite concentrations in all samples were low (<1 mg/L,Table 2). For the samples from WWT"Ps which have high ammonia concentrations, partial nitrification to nitrite was expected. Final effluent samples were low because of full nitrification in the during the treatment processes. Corresponding to nitrite concentrations, nitrate concentrations were high for final effluent samples. They show the same trend as the data for the Fargo WWTF (Simsek et al, 2012; Liu, et al., 2013).

Table 2. Concentration (mg-N/L) of Dissolved Nitrogen in Different locations, (average ±SD)

	ZTP	ITP	MTP
DNO_3-N	6.81 ± 2.40	6.33 ± 3.80	7.83 ± 6.49
DNO_2-N	0.48±0.040	0.70±0.048	0.013±0.002
DNH_4-N	0.039±0.0030	0.70±0.48	0.06±0.019
DTN	19.30±6.60	41.8±3.39	33.21±5.01

Dissolved organic Nitrogen (DON)

Due to the difficulties associated with the measurement of DON, there is a shortage of information about the structure and behavior of wastewater- derived DON. By measuring DON and nitrogen containing compounds combined with DON characterization, we have gained new insight into the nature and properties of wastewater-derived DON.

The results show that the average concentration was ranged from 11.71 to 34.82 (mg - N/L). The highest concentration of DON was noticed in ITP sample with average 34.82±3.68 (mg/L ± SD) and the lowest concentration was detected in ZTP sample with average 11.71±1.16 (mg/L ± SD) (Figure 2).

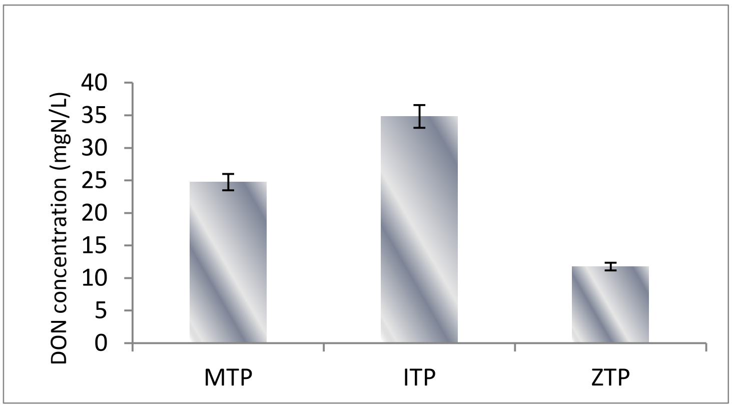

Figure 2. Average DON profile from three WTTPs.

The differences in the concentrations of DON were suggested to be nitrogenous organic compounds could be removed during transportation through the ditch canals by biological processes. The nitrogen present in wastewater is primarily in proteins and urea. Decomposition readily changes the organic form to ammonia. The age of wastewater is indicated by the relative amount of ammonia that is present (Metcalf and Eddy, 2003).

The Figure 3 represented the proportion of DON to TDN in different three WWTPs with different time. The data revealed that the percentage of DON varies with the time of collection and the highest percentage found in the spring in the ITP and MTP while in ZTP in winter was significantly appear (Figure 3). The results shown that the averages of the proportion of the DON were 76.3, 75.2 and 56.75 in the ITP, MTP and ZTP respectively.

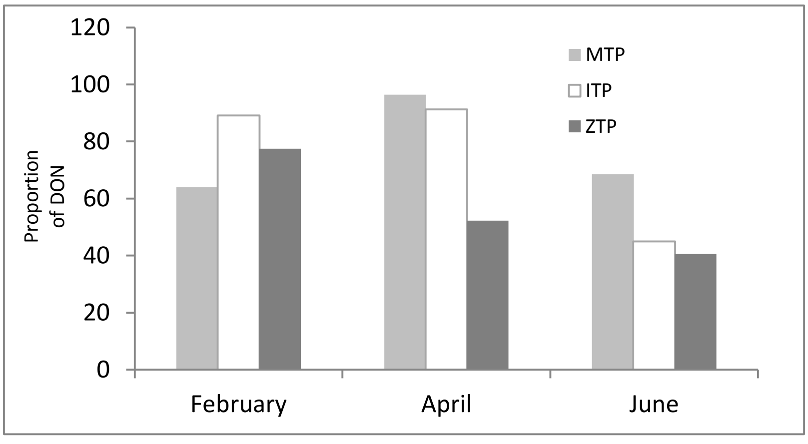

Figure 3. Represent the proportion of DON to TDN in different three WWTPs with different time.

The data revealed that the treatment plant removed of the DON. Similar to inorganic nitrogen removal, major removal of DON around 43.3% was observed in the ZTP which mainly operated by the biological processes of the plant. DON fractions of TDN were 24% and 25% in the plant effluent of ITP and MTP, respectively. The results agreed with the previous observation (Huo, et al., 2013)

Figure 4 shown that the concentrations of DIN were lower than the DON with average 7.01±4.04, 8.43±6.56 and 7.58±1.75 mg/l, (ZTP, MTP and ITP, respectively). To compare the concentrations of DIN in different plants with the concentrations of DON it found that the mechanisms in the plants tried to remove a large amount of DIN fraction compare to DON fraction.

DIN and DON from three WTTPs.

In addition, all dissolved nitrogen species were monitored in the three WWTPs and the results shown in the Figure 5. It was found that there was very little or no nitrite in the samples. the dissolved nitrogen concentration ranged from 0.3 to 7.8 mg N/L. Nitrate was the dominant species and was approximately 8-21% of dissolved nitrogen.

The most variable plant is the MTP, which comprised of up to 24 percent of influent N as ON. Similar results were reported by Pagilla et al. (2008) in the samples from 3 fully nitrified WWTPs in Illinois and 4 BNR plants in Poland. The fraction of influent ON ranged from 2 to 21 % and 13 to 27% in Illinois and Polish plants, respectively. In high influent ON concentration, the results show that ON concentration decreased during the primary treatment process and along the treatment processes. This could be due to dilution from recycle flows or biological processes when wastewater was exposed to treatment conditions in preliminary and primary treatment processes.

Ammonium removal was mainly observed in biological process. The results showed that all WWTPs efficiently removed ammonium and nitrate was present in very low concentration in the effluent. It was found that complete nitrification was achieved. The similarity was found in all 3 plants.

Biodegradable dissolved organic nitrogen BDON

Out of the initial DON of 3.18 mg N/L, nearly 1.95 mg N/L (30% DON bioavailable) was bioavailable to algae at the end of the 14-day bioassay as seen by the net DON concentration reduced over the 14-day period (Fig 6). The biodegradability/bioavailability of DON drops significantly. The removal percentage of the DON was increased during the biodegradation process so the removal BDON was about 30% within 14 days (Figure 6). The BDON degradation rate constant was found to be 0.03/day.

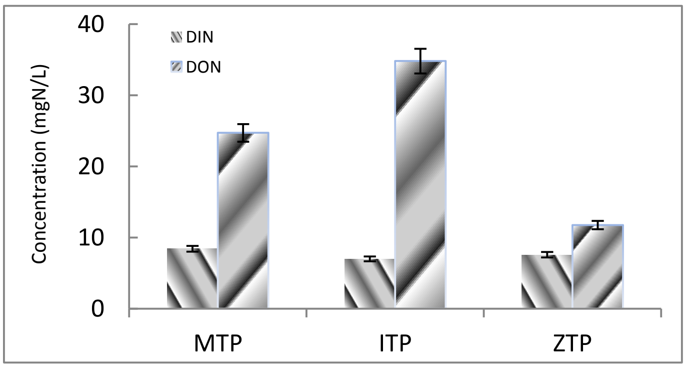

Figure 4. represented the comparison between the concentrations of DIN and DON from three WTTPs.

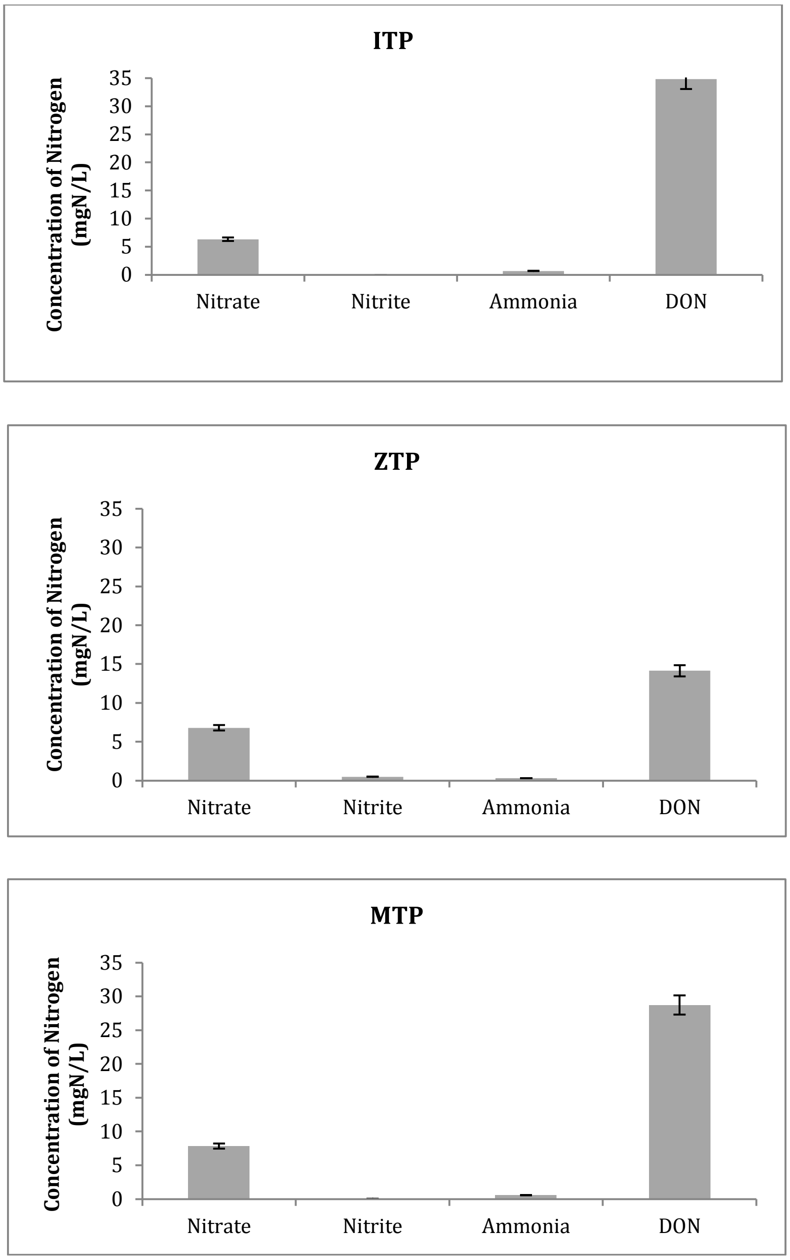

Figure 5 . Summary nitrogen species concentrations in the studied WWTPs.

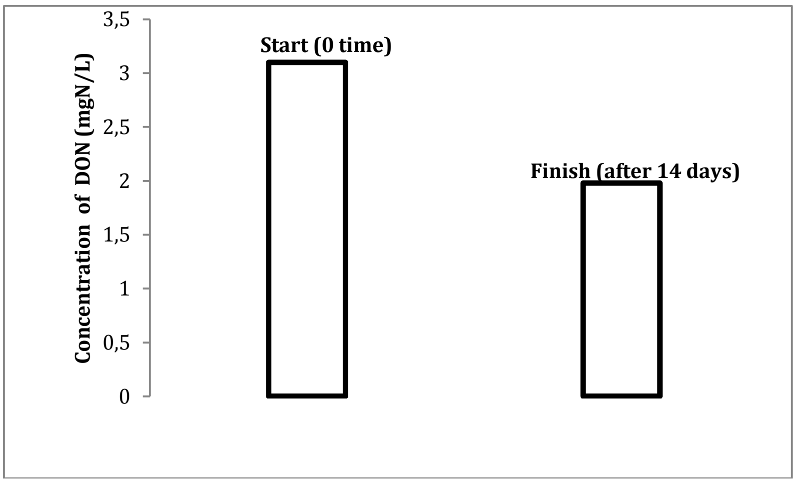

Figure 6. Represent the concentration of BDON from the first day to 14 days of processing

The results from DON bioavailability experiments showed that DON derived from WTPs is bioavailable either directly or indirectly to algae as seen from decrease in DON concentration, at the end of the 14-day incubation period.

Similar trends of BDON concentrations along treatment train were also reported for e US WWTPs (Czerwionkaet al 2012).These studies showed that a portion of effluent DON was biodegradable and/or bioavailable regardless of the type of wastewater treatment processes (Bronk, et al., 2010), and a significant portion of effluent DON was difficult to remove during biological treatment (Pehlivanoglu-Mantas and Sedlak 2008). The information of DON profile along treatment train would help to understand the roles of wastewater treatment process in the removal of this fraction of DON (Simsek et al, 2012).

As a conclusion the results were provide insight into the nature of wastewater-derived DON in some Jordanian WWTPs. The total DON account approximately for more than 70% of the TDN from the effluent leaving WWTP. Beside the dissolved amino acids 70% of the DON unidentified. It is likely that these compounds consist of a complex suite of partially metabolized compounds of biogenic origin. Although it may not be possible to chemically characterize, and identify all the individual nitrogen-containing compounds.

ACKNOWLEDGMENT

This research was supported by Deanship of Research/ The Hashemite University. We thank the technician in our lab for assistance for analyses, and for comments that greatly improved the manuscript.

REFERENCES

American Public Health Association. Water Environment Federation (APHA). (1998) Standard Methods for the Examination of Waters and Waste waters,.

Avnimelech, Y., Mozes, N., Diab, S., & Kochba, M. (1995). Rates of organic carbon and nitrogen degradation in intensive fish ponds. *Aquaculture, 134*(3), 211-216.

Bataineh, F., Najjar, M., &Malkawi, S. (2002) *Wastewater Reuse, in water demand management forum.* Amman, Jordan

Bronk D A, Roberts Q N, Canuel E A, Mesfioui R, Filippino K C, Mulholland M R and Love N G (2010). Effluent organic nitrogen bioavailability and photochemical and salinity-mediated release. *Environ. Sci. Technol. 44.* 5830–5.

Bronk, D. A., See, J. H., Bradley, P., &Killberg, L. (2007). DON as a source of bioavailable nitrogen for phytoplankton. *Biogeosciences, 4*(3), 283-296.

Czerwionka K, Makinia J, Pagilla K R and Stensel H D (2012). Characteristics and fate of organic nitorgen in municipal biological nutrient removal wastewater treatment plants. *Water Res. 46,* 2057–66.

Huo, S., Xi, B., Yu, H., Qin, Y., Zan, F., & Zhang, J. (2013). Characteristics and transformations of dissolved organic nitrogen in municipal biological nitrogen removal wastewater treatment plants. *Environmental Research Letters, 8*(4), 044005

Lee W and Westerhoff P (2005). Dissolved organic nitrogen measurement using dialysis pretreatment. *Environ. Sci. Technol. 39,* 879–84.

Lee W, Westerhoff P and Croues J P (2007). Dissolved organic nitrogen as a precursor for chloroform, dichloroacetonitrile, N-nitrosodimethylamine, and trichloronitrometha. *Environ. Sci. Technol. 41,* 5485–90.

Liu H Z, Jeong J, Gray H, Smith S and Sedlak D L (2012). Algal uptake of hydrophobic and hydrophilic dissolved organic nitrogen in effluent from biological nutrient removal municipal wastewater treatment systems. *Environ. Sci. Technol. 46*, 713–21.

Metcalf, E. (2003). Inc., *Wastewater Engineering, Treatment and Reuse*. New York: McGraw-Hill.

Pagilla K R, Czerwionka K, Urgun-Demirtas M and Makinia J (2008). Nitrogen species in wastewater treatment plant influents and effluents-the US and Polish case studies. *Water Sci. Technol*. *57,* 1511–7.

Pagilla K R, Urgun-Demirtas M and Ramani R (2006). Low effluent nutrient technologies for wastewater treatment. *Water Sci. Technol. 53*, 165–72.

Pehlivanoglu-Mantas E and Sedlak D L (2004). Bioavailability of wastewater-derived organic nitrogen to the alga Selenastrum Capricornutum. *Water Res. 38*, 3189–96.

Pehlivanoglu-Mantas E and Sedlak D L (2006). The fate of wastewater-derived NDMA precursors in the aquatic environment. *Water Res. 40,* 1287–93.

Pehlivanoglu-Mantas E and Sedlak D L (2008). Measurement of dissolved organic nitrogen forms in wastewater effluents: concentrations, size distribution and NDMA formation potential. *Water Res. 42,* 3890–8.

Sattayatewa C, Pagilla K, Pitt P, Selock K and Bruton T (2009). Organic nitrogen transformations in a four-stage Bardenpho nitrogen removal plant and bioavailability/biodegradability of effluent DON. *Water Res. 43,* 4507–16.

Simsek H, Kasi M, Ohm J B, Blonigen M and Khan E (2013). Bioavailable and biodegradable dissolved organic nitrogen in activated sludge and trickling filter wastewater treatment plants. *Water Res. 47,* 3201–10.

Simsek H, Kasi M, Ohm J B, Wadhawan T, Bye C, Blonigen M and Khan E (2012). Fate of dissolved organic nitrogen in two stage trickling filter process. *Water Res. 46*, 5115–26

Vandenbruwane J, Neve S D, Qualls R G, Salomez J and Hofman G (2007). Optimization of dissolved organic nitrogen (DON) measurements in aqueous samples with high inorganic nitrogen concentrations. *Sci. Total Environ. 386,* 103–13.

Xu B, Li D P, Li W, Xia S J, Lin Y L, Hu C Y, Zhang C J and Gao N Y (2010). Measurements of dissolved organic nitrogen (DON) in water samples with nanofiltration pretreatment *Water Res. 44,* 5376–84.

◈◇◈