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Temperature sensitivity of nitrate removal in woodchip bioreactors increases with woodchip age and following drying-rewetting cycles

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Water Impact Statement

Results indicated temperature sensitivity of nitrate removal in woodchip bioreactors is related to short and long-term changes in carbon quality, providing an alternative hypothesis for these changes seen in a previous study on woodchip bioreactors. Declines in nitrate removal efficiency were greatest at lower temperatures, vital information for the design of these sustainable systems to achieve nutrient reduction goals in cold climates.

Abstract

Woodchip bioreactors are a beneficial management practice with increasing use for the sustainable reduction of nitrate in waters discharged from agriculture and urban landscapes. Previous research has shown an interaction between temperature and carbon quality with respect to microbial respiration, which may affect performance of woodchip bioreactors. This study used two previously published data sets of woodchip bioreactors in Spain and the United States that were exposed to weekly drying-rewetting cycles, to better understand the processes driving changes in temperature sensitivity of nitrate removal. The factor by which nitrate removal increased given a 10 °C increase in temperature (Q_{10}) was used as a metric for temperature sensitivity. . Values of Q_{10} for nitrate removal in both experiments ranged from 1.8 – 3.1 and generally increased over time as woodchips aged. In field bioreactors, mean nitrate removal rate at temperatures 10 – 15 °C and 22 – 27 °C decreased by 36% and 7%, respectively, from the first to second year. Values of Q_{10} increased with amount of time since resaturation of the woodchips following a drying-rewetting cycle. Dynamic calculations of Q_{10} showed changes in Q_{10} were not unidirectional. Subsetting the datasets showed that Q_{10} was temperature-dependent and varied according to minimum temperature value and total range in temperature. Results suggest temperature sensitivity of nitrate removal was related to short and long-term changes in carbon quality or availability, consistent with the carbon-quality-temperature hypothesis. When sizing woodchip bioreactors, water quality managers should consider that long-term declines in efficiency will be greatest at lower temperatures.

1. Introduction

Woodchip bioreactors, also referred to as denitrification beds or denitrifying bioreactors, are an agricultural Beneficial/Best Management Practice (BMP) used for the removal of nitrate (NO_3^-) in water discharged from agriculture. When NO_3^- is the predominant nitrogen (N) species, the primary removal process is denitrification where N in the aqueous NO_3^- anion form is reduced

into gaseous dinitrogen and nitrous oxide. In woodchip bioreactors, denitrifying conditions are favored by providing a carbon substrate (i.e., woodchips) as the electron donor in the anaerobic respiratory pathway. Prolonged anoxic conditions are maintained to favor use of NO_3^- as the electron acceptor. These systems are attractive both for their high drainable porosity (60 – 80%) [1,2], low cost and maintenance needs, and comparatively long lifespan (~10 – 15 years) as lignocellulosic woody material degrades slowly relative to more labile carbon sources.

Factors influencing NO_3^- removal efficiency in woodchip bioreactors include temperature [3 – 5], hydraulic residence time (HRT) [4, 6, 7], influent NO_3^- concentration, and age of woodchips [3, 8, 9]. Temperature is known to affect microbial metabolic activity (e.g. denitrification) through increased metabolic rates at higher temperatures [10 – 12] and is seen in increased NO_3^- removal in woodchip bioreactors at higher temperatures [3, 5, 9]. The relationship between NO_3^- removal rates and temperature has been quantified using the Q_{10} temperature coefficient. The Q_{10} coefficient corresponds to the factor by which NO_3^- removal rates increase for every 10 °C increase in temperature, with $Q_{10} = 1$ indicating no temperature effect, and higher Q_{10} values indicating greater sensitivity to temperature. Reported Q_{10} values for NO_3^- removal in woodchip bioreactors typically range from 1.8 – 4.7 [5, 13 – 15]. Unrelated to temperature, NO_3^- removal rates in woodchip bioreactors are also known to generally decrease with time. As woodchips age, a higher proportion of the biomass is comprised of lignin [16], relative to hemicellulose and cellulose which are more rapidly consumed or leached, with low or negligible rates of consumption of the more recalcitrant lignin via anaerobic respiratory pathways [17 – 19]. Increasing recalcitrance of the wood-derived carbon is believed to be the cause of decreased NO_3^- removal rates over time [4, 9], with most of this decrease occurring during the first year as fresh, labile carbon is quickly lost and carbon quality of the woodchips decreases. For clarity, carbon quality here is considered as the number of steps required to fully respire a carbon atom from an organic compound and release it as carbon dioxide [20].

The effects of temperature and woodchip age on NO_3^- removal in woodchip bioreactors has generally been determined by quantifying their impact as independent factors. There is evidence, however, of an interaction between the two factors, with temperature effect changing as carbon quality of the woodchips changes over time. Experimental evidence of increased temperature sensitivity of respiration at lower carbon quality has been widely reported [21 – 23]. Xu et al. showed that temperature sensitivity of respiration was inversely correlated with soil organic

carbon quality, with higher Q_{10} at lower carbon quality [24]. It was recently shown that Q_{10} values of NO_3^- removal rates in laboratory pine-woodchip bioreactors increased over a 480 d period [25]. While the authors agreed the increase in Q_{10} was likely tied to a decrease in carbon quality of the woodchips, it was further proposed that the temporal change in Q_{10} was due to increased activity of fermenting bacteria. The authors contended that denitrifying bacteria are dependent on substrates (i.e., fermentation by-products) produced by upstream fermenting bacteria, and it was changes in cross-feeding between fermenting and denitrifying communities over time that resulted in the change in Q_{10}.

Drying-rewetting (DRW) cycles have been shown to increase NO_3^- removal rates in woodchip bioreactors [26], with increasing duration of aerobic periods prior to woodchip resaturation leading to greater increases in removal rates [27]. The hypothesized mechanism for this effect was that DRW cycles, by briefly exposing the carbon substrate to aerobic conditions, effectively increase carbon availability by promoting aerobic breakdown. Increased degradation of woodchips more frequently exposed to aerobic conditions was seen by Moorman et al. as greater biomass loss in shallower woodchips [28], and Ghane et al. who showed that woodchips closer to a bioreactor inlet, prior to depletion of dissolved oxygen, had greater proportions of recalcitrant carbon as lignin [16]. Aerobic processes are more capable of degrading lignin [29] and yield lower molecular weight carbon molecules [30, 31] that are more bioavailable to denitrifiers. Carbon leaching from organic material decreases quickly (i.e., within a matter of days) upon resaturation after a DRW cycle [26, 32, 33] as aerobically-produced carbon is leached or consumed. Considering the broadly accepted carbon quality-temperature hypothesis (i.e., that temperature sensitivity of respiration increases with decreasing carbon quality) and that DRW cycles result in short-term increases in carbon quality, there should be observable changes in temperature sensitivity of NO_3^- removal in woodchip bioreactors not only across long-term time scales (i.e., woodchip age over years), but also in relation to short-term dynamics following DRW cycles.

This paper uses two previously published data sets to perform a comprehensive analysis of the relationship between wood age and time since a DRW cycle (i.e., indicators of carbon quality) and temperature and the interaction of the two factors on NO_3^- removal rates in woodchip bioreactors.

2. Materials and Methods

Two published data sets were used to observe the interaction of carbon quality and temperature and its effect on NO_3^- removal rates [26, 27, 34]. The two data sets were derived from separate experiments with markedly different influent water characteristics, experimental procedures, and measurement methods. They are described briefly in the following two sections, and more detailed methods and results can be found in the cited publications. Carbon quality of woodchips or dissolved organic carbon in the bioreactor effluent was not directly measured in either study. Instead, woodchip age and elapsed time since rewetting following a DRW cycle were used as metrics for carbon quality to determine its effect on the temperature sensitivity of NO_3^- removal over short and long-term time scales.

2.1 UPCT - Batch experiments treating concentrated brine

Results from three pilot-scale woodchip bioreactors treating concentrated brine were previously reported (Díaz-García et al., 2019). Experiments were conducted at the Agri-food Experimental Station Tomás Ferro (ESEA) (N 37° 41' 17.6" and W 0° 57' 04.4") of the School of Agricultural Engineering of Universidad Politécnica de Cartagena (ETSIA-UPCT) in Cartagena, Region of Murcia, Spain. Three rectangular tanks (142 x 109 cm) were filled with chopped, citrus woodchips (depth 85 cm) obtained from agricultural sources in the region. Influent water consisted of reject brine (electrical conductivity 16 – 20 mS cm^{-1}, influent NO_3^- concentration = 38 – 59 mg N L^{-1}) from a desalination plant providing irrigation water, with water sourced from an adjacent brackish aquifer contaminated with NO_3^- from fertilizer and other salts (e.g. Mg^{2+}, Ca^{2+}, Na^+, Cl^-) from seawater intrusion. Bioreactors were located at the open-air facility and were therefore exposed to daily and seasonal changes in temperature. The experiment and the data obtained from it is subsequently referred to as UPCT.

Batch experiments were performed over 730 days from December 2017 to November 2019. During UPCT batch experiments, woodchip bioreactor tanks were completely-filled with brine (200 – 330 L) until water level was even with the woodchip surface. Woodchips remained fully saturated for 24 h during each batch, after which tanks were completely drained and effluent samples collected. Once the brine was removed from the bioreactors, they were immediately refilled (<1 h after drain) and woodchips resaturated with new brine for the next 24 h batch experiment. A single batch refers to the 24 h period in which woodchips were saturated with

untreated reject brine from reverse osmosis, and the denitrified brine later emptied after 24 h. Over the entire 730 d experiment, three batch experiments were performed each week beginning on Monday of each week. Following the third 24 h batch experiment of each week on Wednesday, no water was added to the bioreactors and woodchips remained unsaturated for a period of 96 h until the first batch on Monday of the following week, constituting the DRW cycle for this experiment. Data collected from first, second, and third batch runs of the week are referred to as Batch 1, Batch 2, and Batch 3, respectively.

Influent and effluent samples were collected, respectively, before and after each batch, filtered through 0.45 μm size filter (Sartorius GmbH) prior to analysis. The samples were analyzed for NO_3^- concentration using double channel chromatographic system 850 Professional Ion Chromatography Metrohm at the SAIT-UPCT analytical lab in Cartagena. Concentrations of N species are reported in terms of mass nitrogen (i.e., mg N L^{-1}). Water temperature inside the bioreactors was measured using a Hanna handheld data logger (HI98194) with a pH/EC/temperature multiparameter probe (HI7698194) by dipping the probe into a PVC porewater well (6.3 cm) until a stable reading was reached. Batch experiments began in the early morning (t = 0 h) and finished the following morning (t = 24 h), with variable temperatures observed over each 24 h batch. Temperature values were an average of measurements taken at 30 min, 10 h and 24 h after filling the bioreactors, giving a daily average. Although diurnal temperature changes would affect microbial activity throughout the day, our aim was not to evaluate this effect but the effect of annual temperature variation (i.e., seasonal), on basis of the average daily temperature.

2.2 NCSU – Continuous flow in lab column study

The second data set used in this study was obtained from two separate lab experiments done at North Carolina State University (NCSU) investigating the effect of DRW cycles on NO_3^- removal in woodchip bioreactors [26, 27]. In both lab experiments, eight woodchip-filled columns (15 cm diameter x 95 cm height) were operated in continuous flow. Columns were first monitored in 2017 over a period of 287 d [26] during which columns received continuous flow (HRT = 8±1 h, mean±standard deviation) from a stock tank of dechlorinated tap water dosed with KNO_3 (influent NO_3^- concentration = 19.6±1.3 mg N L^{-1}). A follow-up, 108 d experiment in 2018 [27] used the same columns with similar flow rates and influent NO_3^- concentration as the

2017 experiment (HRT = 8±1 h; influent NO_3^- concentration = 17.1±0.3 mg N L^{-1}). The two experiments and the data obtained from them are jointly referred to as NCSU.

In the first NCSU experiment (2017), a total of eight woodchip-filled columns were used. One treatment consisted of constant saturation (SAT) of the woodchips provided by continuous and uninterrupted upflow in four of the eight columns throughout the entire experiment. Water level in the SAT columns remained constant at the level of the column outflow, the upper surface of the woodchip media. The second treatment, performed in the other four columns, consisted of exposing the woodchips to unsaturated conditions for 8 h once a week in weekly drying-rewetting cycles (DRW) as follows; flow to DRW columns was stopped once a week by disconnecting the inflow lines, after which the DRW columns were drained rapidly (~15 min time to drain) and left unsaturated for 8 h, exposing the woodchips to unsaturated conditions. After this 8 h period where woodchips were unsaturated, flow to DRW columns was reestablished by reconnecting the inflow line. The second NCSU experiment in 2018, beginning 163 days after the end of the 2017 experiment, used four of the same columns from the prior experiment, applying the SAT and 8 h DRW treatments to two columns each. Columns reused in the 2018 NCSU experiment received the same treatment they were given in the 2017 experiment (i.e., two of the SAT columns from 2017 were also given SAT conditions in 2018). A total of 39 and 11 weekly 8 h DRW cycles were applied to the DRW treatment in 2017 and 2018, respectively. Woodchips were 558 d in age by the end of the 2018 NCSU experiment.

In both NCSU experiments, stock tank and column outflow water chemistry were measured using a small volume multiplexed pumping system (MPS) [35] coupled to a high frequency spectrophotometer. The MPS sequentially pumped 25 mL samples from each column for absorbance measurement by a field spectrophotometer (Spectro::lyser; manufactured by s::can, Type SP-1-035-p0-s-NO-075) fitted with a 4 mm pathlength, 1.1 mL flow through quartz cuvette (46-Q-4, Starna Cells, Inc.). Concentrations of NO_3^- in the stock tank and outflow of each column were measured on 2 h intervals. Nitrate concentrations were calculated from the absorbance measured by the spectrophotometer following methods previously described [36, 37]. For improved accuracy of the spectrophotometer, an experiment-specific calibration was used rather than the manufacturer's calibration. Sample volumes analyzed by the spectrophotometer were submitted for lab analysis (EPA Method 353.2, BAE Environmental Analysis Lab, North Carolina State University) to calibrate the probe for NO_3^- and DOC. In the 2017 NCSU experiment, column outflow was monitored only during Days 0 – 98, 147 – 171, and 252 – 287, although columns received continuous upflow over the entire 287 days. In the 2018 NCSU experiment, column outflow was monitored over the full duration of the 108 d experiment.

Temperature of column outflow was measured hourly using Presens® temperature sensors (DP-PSt3, Presens Precision Sensing GmbH). Temperature sensors were inserted through the top of the column and placed such that the sensor tips were at least 2 cm below the surface of woodchip media, per manufacturer's specifications. Water temperature measurements were made on an hourly interval.

2.3 Nitrate Removal Rates

Hydraulic loading of woodchip bioreactors differed between the UPCT and NCSU experiment. Data obtained from the UPCT experiment reflect performance of bioreactors run in batch, while NCSU woodchip columns were provided continuous, uninterrupted flow outside of DRW cycles. Methods of calculating volumetric NO_3^- removal rates (R_{NO3}), a commonly reported metric for woodchip bioreactors, were different between experiments. Volumetric rates were calculated according to Equations 1 and 2 for the UPCT and NCSU experiments, respectively:

$$\text{Equation 1.} \quad \frac{([NO_3]_{in} - [NO_3]_{out}) * V_{water}}{t * V_{saturated\ woodchips}}$$

222 Equation 2. $$\frac{([NO_3]_{in} - [NO_3]_{out}) * Q}{V_{saturated\ woodchips}}$$

223

224 where, in Equation 1, $[NO_3^-]_{in}$ and $[NO_3^-]_{out}$ are the NO_3^- concentrations in the initial brine and in
225 the effluent after 24 h, V_{water} is the volume of water added to the woodchips during each batch, t
226 is the duration of time which water was in contact with the woodchips (i.e., 24 h), and $V_{saturated}$
227 $_{woodchips}$ is the volume of saturated woodchips in the rectangular tanks (1.32 m^3). In Equation 2,
228 $[NO_3^-]_{in}$ and $[NO_3^-]_{out}$ were the NO_3^- concentrations measured at the column inlet and outlet
229 every 2 hours, Q was the flow rate at the time of the NO_3^- measurements, and $V_{saturated\ woodchips}$ is
230 the volume of saturated woodchips in the upflow columns (0.009 m^3). Removal rates were
231 reported in units of g N m^{-3} d^{-1}. In this study's analysis, R_{NO3} was used as a metric to reflect the
232 biogeochemical rates of NO_3^- removal. For woodchip bioreactors, it has been generally assumed
233 that denitrification is responsible for the majority of reduction in NO_3^- concentration, rather than
234 other processes such as dissimilatory NO_3^- reduction to ammonium or annamox which also occur
235 under anoxic conditions [38, 39]. This was likely the case in both UPCT and NCSU experiments,
236 since NH_4^+ concentrations in both the influent and effluent were generally less than <2 mg N L^{-1}.
237 In subsequent discussion and analysis, it is assumed that changes in R_{NO3} reflected changes in
238 denitrification rates, although the methods used in both experiments did not directly measure
239 denitrification.

240 **2.4 Temperature sensitivity**

241 Temperature sensitivity of R_{NO3} in both studies was quantified by calculation of the Q_{10} value, or
242 the factor by which a rate increases for every 10° C increase, a common metric used for
243 quantifying temperature sensitivity of a biogeochemical process [25, 40, 41]. Measurements of
244 R_{NO3} during each experiment were matched with corresponding temperature measurements. Data
245 were then fitted to Equations 3 and 4 to calculate Q_{10},

246 Equation 3 $$R_T = R_0 * e^{kT}$$

247 Equation 4 $$Q_{10} = e^{10*k}$$

248 where R_T is the observed R_{NO3} (g N m^{-3} d^{-1}) at a given temperature from measured influent and
249 effluent NO_3^- concentration, R_0 is a constant for the intercept, k is a constant describing the slope

of the temperature relationship, and T is the measured temperature value. Collected data was fitted to the relationship in Equation 3 using the nls() function in R Studio [42], a function finding the least-squares parameter estimates of a nonlinear function, solving for R_0 and k. Data from UPCT and NCSU were analyzed separately.

In the UPCT experiment, the short-term effects of carbon quality on R_{NO3} temperature sensitivity were analyzed by separating data from the first, second, and third batch of each week following the 96 h unsaturated period (e.g. Q_{10} for first day after DRW cycle considered only Batch 1 data). In the NCSU experiment, this short-term effect of carbon quality was analyzed by separating data according to number of days since the weekly, 8 h DRW cycle (e.g. Q_{10} for first day after DRW cycle considered only data from first 24 h of continuous flow following the resaturation of the woodchips). The 2017 and 2018 data for the NCSU experiment were combined to form a single data set. The first 30 days of measurements in UPCT and NCSU experiments were removed from both data sets prior to temperature sensitivity analysis due to high amounts of organic carbon leaching in this initial period (see Section 3.1.1 and 3.2.1). Values of Q_{10} for DOC release were also calculated, substituting effluent DOC concentration into Equations 3 and 4. Standard error of the calculated Q_{10} was included in the analysis, calculated as the change in Q_{10} given by the standard error of the estimate for k in Equation 3. Residual standard error of the model when fitting the data to Equation 3 was used as a measure of goodness of fit.

2.5 Dynamic Q_{10} calculation

Uninterrupted data collection over 730 d during the UPCT experiment provided the opportunity to observe long-term changes in Q_{10} over short time intervals. Q_{10} was calculated dynamically over the 730 d period by subsetting the data according to time, bounded by t_0 and t_1, incrementally advancing the data window by one day at a time. Here, t_0 is the first day of the data window, and t_1 is the final day. Each Q_{10} calculation consisted of 365 d of data, such that t_1 minus t_0 always equaled 365 d (i.e., separate Q_{10} calculations for data collected during Day 30 – 395, 31 – 396, 32 – 397, etc.) The data window was incrementally advanced by one day at a time until t_1 = 730 d. Dynamic Q_{10} was calculated when considering all data combined, and analyzing data from Batch 1, Batch 2, and Batch 3 separately.

2.6 Temperature dependence of Q_{10}

Analysis of the temperature dependence of Q_{10} was performed on the UPCT data set, in which average daily temperatures ranged from 8.9 – 27.8 °C. This was done by subsetting the complete data set at various temperature intervals. Each temperature interval varied by 1) minimum temperature of the interval and 2) range in temperature of the interval. For example, with a minimum temperature of 10 °C and range in temperature of 5 °C, the subsetted data for calculating Q_{10} would contain only measurements from experiments in which temperatures were 10 – 15 °C. For a temperature interval with minimum temperature of 15 °C and range in temperature of 10 °C, the subsetted data would include only measurements from experiments in which temperatures were 15 – 25 °C. Q_{10} was calculated by subsetting the data while varying both minimum temperature and range of the interval at increments of 1 °C. Lowest and highest values for minimum daily average temperature were 10 and 20 °C, respectively, while lowest and highest values of range in temperature were 5 and 15 °C. Q_{10} was not calculated if the temperature interval contained temperatures >25 °C (e.g., 21 – 26 °C or 15 – 27 °C). Data from Days 30 – 395 and 365 – 730 were analyzed separately. Uncertainty of the Q_{10} value was calculated by using the standard error of the k coefficient when fitting the model to Equation 3.

3. Results

3.1 UPCT batch experiments

3.1.1. Organic carbon losses from woodchips

Initial losses of dissolved organic carbon (DOC) were high in both experiments, decreasing rapidly in the first 30 days with slower long-term decreases. In the first three UPCT batch runs, mean DOC concentration in the bioreactors after 8 h was 1567±195, 533±44, 314±45 mg C L^{-1}, respectively (Supplemental Fig. S1). Concentrations of DOC continued to decrease until the 12th batch run, after which point DOC concentrations were relatively stable. High initial flushing of DOC was the reason for excluding data from this period during Q_{10} analysis. Mean DOC concentration after 24 h during the first year was 22.3±10.8 mg C L^{-1}, with lower mean DOC in the second year of 12.1±4.4 mg C L^{-1}. Increased DOC in the effluent was observed at warmer temperatures.

3.1.2. Temperature and R_{NO3} relationships

Over the 730 d UPCT experiment, daily average temperatures ranged from 8.9 – 27.8 °C, with temperatures highest during summer months. Temperature had a clear effect on R_{NO3}, with large variability in R_{NO3} that tracked with seasonal changes in temperature (Fig. 1). R_{NO3} was highest (up to 36.4 g N m^{-3} d^{-1}) during the warmer summer months (24.6±0.9 °C,) and lowest (as low as 7.0 g N m^{-3} d-1) during the colder winter months (12.7±1.7 °C). When considering all data collected from Day 30 – 730, the k temperature constant (Equation 3) was positive and significant ($p<0.001$), with a calculated Q_{10} value of 1.71±0.03 (mean ± standard deviation) and residual standard error of 4.7 g N m^{-3} d^{-1}.

Values of Q_{10} increased over the 730 d experiment (Fig. 1). To observe long-term changes in Q_{10}, data were separated into three periods (representing the first year, middle of the experiment, and second year), each period 365 days in duration such that seasonal temperature variability was captured. Considering data collected from Day 30 – 395 (first year), Q_{10} was 1.25±0.02 with a residual standard error of 3.7 g N m^{-3} d^{-1}. Looking at data over a one-year period during the middle of the experiment, from Day 110 – 475 (first to second year), Q_{10} increased to 1.51±0.03 with a higher residual standard error of 4.3 g N m^{-3} d^{-1}. In the final year of the experiment, Day 365 – 730 (second year), Q_{10} increased even further to 1.71±0.03 with the lowest residual standard error of 3.0 g N m^{-3} d^{-1}. Changes in R_{NO3} over time were most noticeable at lower temperatures. During these three periods, shown in Fig. 1, mean R_{NO3} at temperatures 10 – 15 °C were 21.3±5.1, 16.1±5.0, and 13.7±3.2 g N m^{-3} d^{-1}, respectively. There was less variation in mean R_{NO3} at higher temperatures (22 – 27 °C), with values of 27.2±2.7, 27.2±2.7, and 25.4±2.4 g N m^{-3} d^{-1}, respectively. Mean R_{NO3} at 10 – 15 °C during Days 365 – 730 (second year) decreased by 36%, relative to Days 30 – 395 (first year), while mean R_{NO3} at 22 – 27 °C decreased by only 7%.

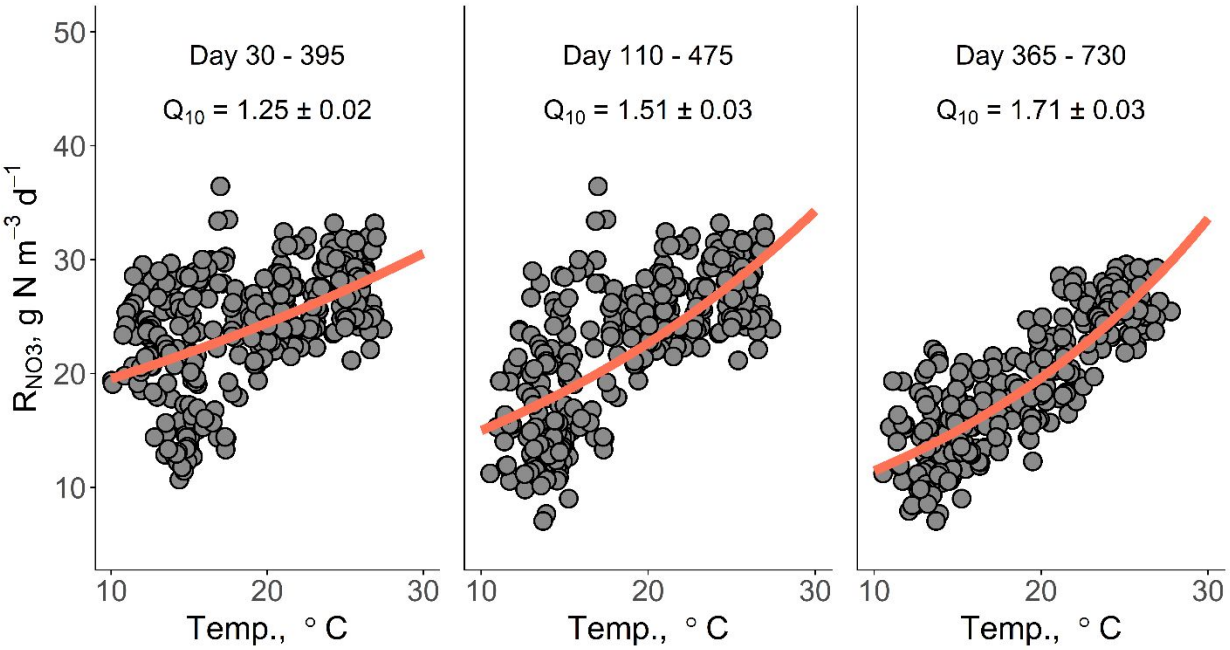


Fig. 1. Relationship of volumetric NO_3^- removal rates, R_{NO3}, with temperature during Day 30 – 395 (first year), Day 110 – 475 (first to second year), and Day 365 – 730 (second year) along with calculated Q_{10} values (estimate ± standard error) in UPCT bioreactors. Calculated Q_{10} increased over the course of the experiment, largely driven by lower R_{NO3} at low temperatures as time increased.

3.1.3. Effects of drying-rewetting cycles

In the UPCT experiment, Q_{10} increased with increasing number of days following the DRW cycle. It should be remembered that for the UPCT bioreactors, woodchips were exposed to 96 h of unsaturated conditions following the last batch of the week (Batch 3), with Batch 1, 2, and 3 occurring on the first, second and third day following resaturation of the woodchips. In the first year (Day 30 – 395, Fig. 2, black solid circles), Q_{10} was lowest for Batch 1 (1.18±0.02) and greatest for Batch 3 (1.34±0.03). Change in Q_{10} from Batch 1 to Batch 2 (0.08) was comparable to the change from Batch 2 to 3 (0.08). The same trend was seen in Day 365 – 730 (second year, Fig. 2, hollow triangles). Batch 1 saw the lowest Q_{10} (1.35±0.03) with a greater difference between Batch 1 and Batch 2 (0.55). The highest Q_{10} was in Batch 3 (2.01±0.06). For all batches, Q_{10} was greater in the second year, although the largest Q_{10} increases from the first to second year were for Batch 2 (0.64) and Batch 3 (0.67). Residual model errors for each batch were higher in the first year, at 3.2, 3.1 and 3.6 g N m^{-3} d^{-1} for Batch 1, 2, and 3, respectively. Residual errors in the second year decreased to 2.6, 2.4, and 2.1 g N m^{-3} d^{-1}.

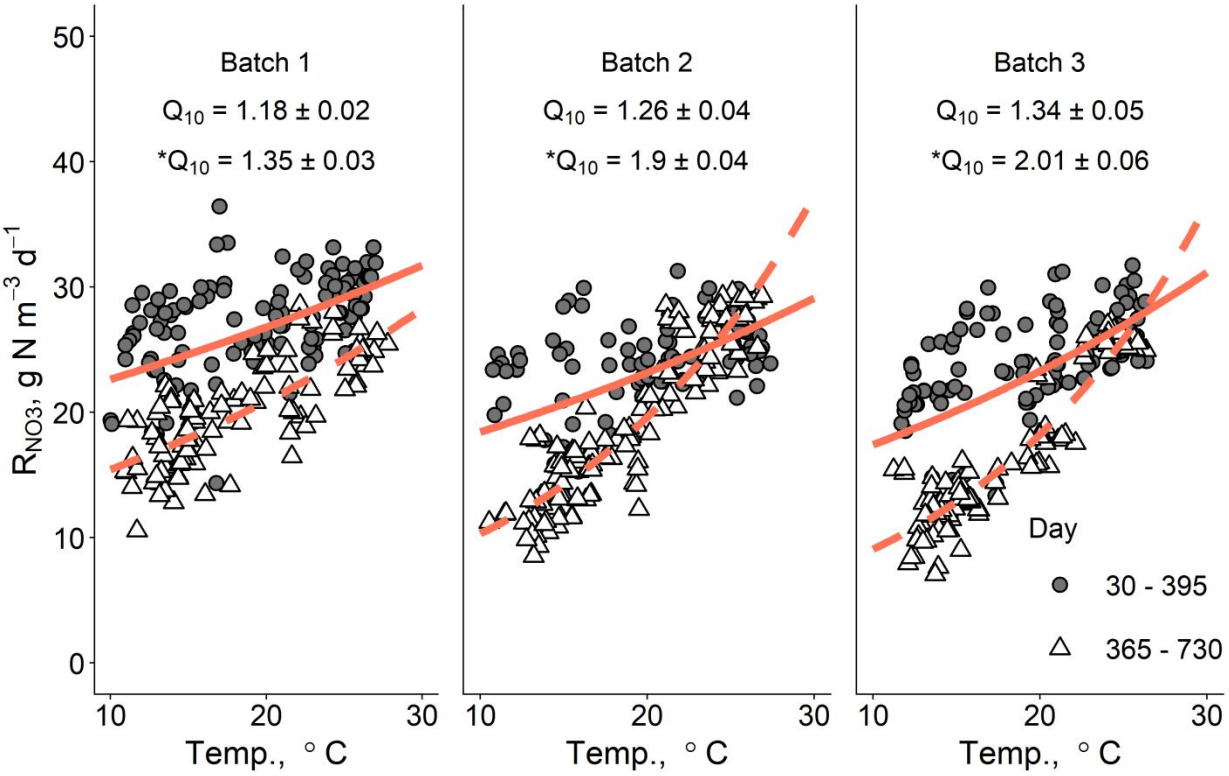
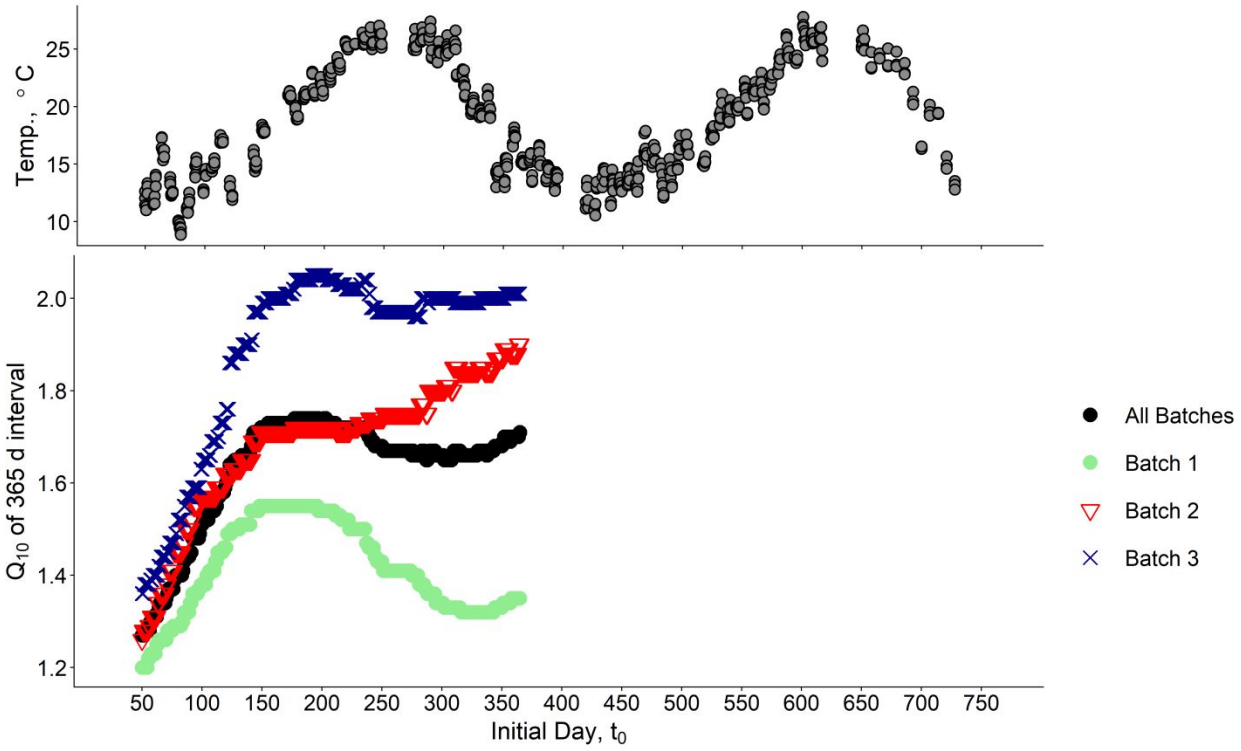


Fig. 2. Relationship of volumetric NO_3^- removal rates, R_{NO3}, with temperature calculated for each batch run of the week in UPCT bioreactors during Days 30 – 395 (first year, black circles, solid line) and Days 365 – 730 (second year, hollow triangles, dashed line). Q_{10} values for Days 365 – 730 are denoted by the asterisk (*). In both periods, Q_{10} increased with time since the DRW cycle, with higher Q_{10} during the second year for all batches.

3.1.4. Dynamic Q_{10} calculations

Calculated Q_{10} based on data from all batches increased quickly at the beginning of the experiment from 1.25 – 1.73 over Days 50 – 155 (Fig. 3). The Q_{10} was relatively stable over Days 155 – 210, after which a slight decrease occurred. A similar initial increase over Days 50 – 150 was seen for Q_{10} calculated for Batch 1, Batch 2, and Batch 3. From Days 150 – 210, Q_{10} in both Batch 1 and Batch 2 were relatively stable at 1.54 and 1.72, respectively, although Q_{10} for Batch 3 continued to increase slowly over Days 150 – 200. After Day 210, Q_{10} for Batch 1 decreased until Day ~320, reaching a minimum of 1.32, before increasing again. Q_{10} for Batch 2

364 began increasing on Day ~230, with the highest value of 1.90 on Day 365.



365

366 **Fig. 3.** Q_{10} values calculated for all batches and each batch separately for the 730 d UPCT field
367 experiment. Q_{10} was calculated dynamically over time by advancing the initial day, t_0, of the 365
368 d time window by one day at a time (i.e., Q_{10} value at Day 50 on x-axis calculated using data
369 from Days 50 – 415). Shape and color denote data from all batches or Batches 1, 2, or 3. Q_{10}
370 was not calculated after t_0 = Day 365 since the interval was restricted to a minimum length of
371 365 d. Temperature shown in the upper panel.

372 **3.1.5. Temperature dependence of Q_{10}**

373 Subsetting the data according to temperature intervals showed variation in Q_{10} values (Fig. 4) as
374 minimum temperature (x-axis) and range of the interval (y-axis) varied at 1 °C increments.
375 During Days 30 – 395 (first year) values of Q_{10} ranged from 1.05 – 1.51, excluding a single
376 higher calculated value of 1.85 in subsetted data at 12 – 17 °C (Fig. 4A). During Days 365 – 730
377 (second year) values of Q_{10} ranged from 1.32 – 2.05 (Fig. 4B). In both years, at a minimum
378 temperature of 10 °C (left-most columns of tile plots), Q_{10} increased as range of the temperature
379 interval (y-axis) increased; Q_{10} was 1.05 and 1.33 at 10 – 15 °C (most bottom left tile) in the first
380 (Fig. 4A) and second (Fig. 4B) year, respectively, and 1.15 and 1.79 at 10 – 25 °C (most top left
381 tile) in the first and second year. Uncertainty of the Q_{10} value (calculated using the standard error
382 of the k coefficient when fitting the data to Equation 3) was higher at smaller ranges in

temperature (Supplemental Fig. S2). For example, from Day 30 – 395, uncertainty of the Q_{10} was 5.3 – 16.5% when range of the temperature interval was 5 °C, but uncertainty was < 3% when range of the temperature interval was greater than 13 °C. In both years, uncertainty of the Q_{10} value was <5% when range of the temperature interval was ≥10 °C. Considering the overall Q_{10} values shown in Fig. 1 over the same time periods, analysis of the temperature dependence of Q_{10} showed that Q_{10} varied by up to 48 and 23% in the first and second year, respectively, depending on the temperature interval used.

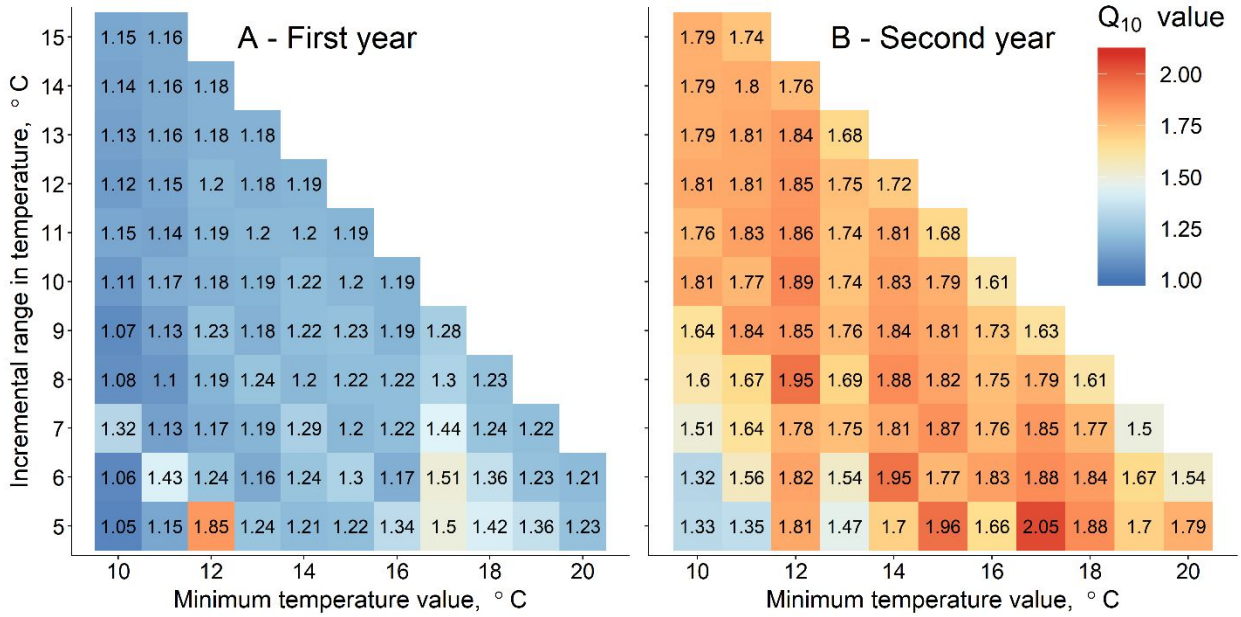


Fig. 4. Tile plots illustrating calculated Q_{10} values for the UPCT field bioreactors during Days 30 – 395 (first year, A) and 365 – 730 (second year, B). Each tile represents a separate Q_{10} value when subsetting the data at various intervals according to minimum temperature (x-axis) and range in temperature of the interval (y-axis). Numbers shown within each tile are the Q_{10} value at the given interval.

3.2. NCSU column study

3.2.1. Organic carbon losses from woodchips

Concentrations of DOC were initially high in effluent from the NCSU columns, although values were much lower relative to UPCT batches since columns were operated in continuous flow with an ~8 h HRT. From Day 20 – 50, effluent DOC concentration was 3.4±0.7 and 3.5±0.7 mg C L^{-1} for SAT and DRW columns, respectively (Supplemental Fig. S3). From Day 50 – 176 mean DOC was 2.8±0.3 and 3.0±0.4 mg C L^{-1} for SAT and DRW columns, and decreased further during Day 252 – 287 to 1.5±0.1 and 1.7±0.2 mg C L^{-1}. During 2018 (Day 480 – 558), mean

DOC was 1.7±0.3 and 2.0±0.4 mg C L^{-1}. Concentrations of DOC were marginally higher in DRW columns, relative to SAT, with the greatest different in DOC concentration immediately following the DRW cycle. In terms of volumetric rates of DOC release, calculated similarly to R_{NO3} using Equation 2, mean rates of DOC release during Day 30 – 287 (2017) were 1.3±0.7 g C m^{-3} d^{-1}, and 1.8±0.9 g C m^{-3} d^{-1} during Day 480 – 558 (2018).

3.2.2. Temperature and R_{NO3} relationships

From Day 30 – 287 temperatures ranged from 18.6 – 29.0 °C (21.6±1.9 °C), while temperatures from Day 480 – 558 ranged from 20.5 – 24.7 °C (22.7±0.9 °C). Temperature had a clear effect on R_{NO3} when considering data from Day 30 – 287 (2017) and 480 – 558 (2018) separately, with the k temperature constant (Equation 3) significant ($p<0.001$) and positive during both periods. When considering all data collected from Day 30 – 558 (2017 and 2018), there was a calculated Q_{10} value of 1.95±0.02 and residual standard error of 3.9 g N m^{-3} d^{-1}.

Unlike the analysis for the UPCT bioreactors, which had uninterrupted data collection over the entire 730 d period, long-term changes in Q_{10} of the NCSU woodchip columns were analyzed by breaking the data into two periods only, the 2017 and 2018 portions of the NCSU experiment (each containing only 287 and 108 d of data collection, respectively). Values of Q_{10} decreased over the 558 d duration of the NCSU experiment (Fig. 5). Lower Q_{10} was seen from Day 30 – 287, relative to Day 480 – 558, and Q_{10} values were not significantly different between the SAT and DRW treatments. Values for Q_{10} were higher during Day 480 – 558, with a larger difference in Q_{10} between the two treatments. Increase in Q_{10} from Day 30 – 287 to Day 480 – 558 was higher for the DRW treatment (0.71) relative to the increase for the SAT treatment (0.25). Residual standard error of the Q_{10} model from Day 30 – 287 was 3.3 and 3.8 g N m^{-3} d^{-1} for SAT and DRW columns, respectively, and 3.3 and 2.6 g N m^{-3} d^{-1} from Day 480 – 558.

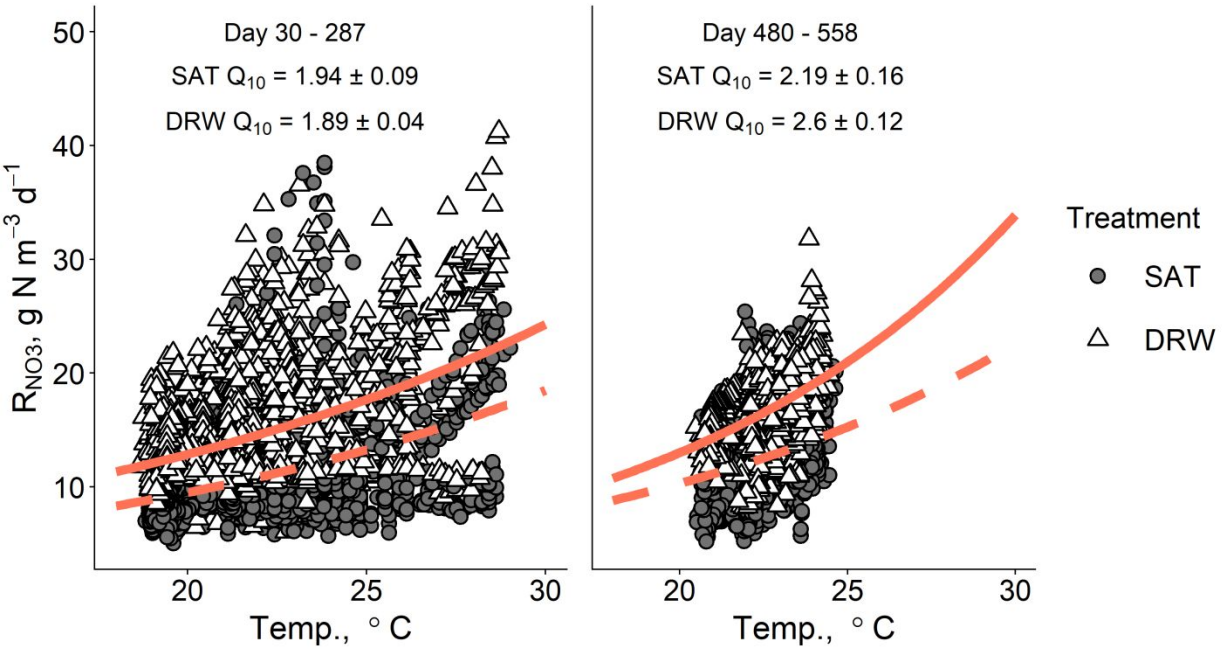


Fig. 5. Relationship of volumetric NO_3^- removal rates, R_{NO3}, with temperature during Day 30 – 287 (2017) and Day 480 – 558 (2018) for the NCSU column experiment. Q_{10} values (estimate ± standard error) were calculated separately for SAT (dashed line) and DRW (solid line) treatments.

3.2.3. Effects of drying-rewetting cycles

Short-term increases in Q_{10} were seen in the NCSU experiment (Fig. 6) when selecting R_{NO3} and calculating Q_{10} separately for each day following the resaturation of the woodchips. Data were not divided by year in this analysis, and data from SAT columns were not used since the columns did not undergo a DRW cycle. In general, Q_{10} increased following the weekly 8 h DRW cycle. A large increase in Q_{10} was seen between Day 1 and Day 2 after rewetting (0.56) and between Day 3 and Day 4 (0.86). Daily increases in Q_{10} were seen in every day until Day 5 following the DRW cycle, with a small decrease in Q_{10} on Day 6. A wider range in temperature for Days 4 – 6 (18.6 – 28.7 °C) after rewetting (Fig. 6), relative to Days 1 – 3 (18.9 – 26.8 °C), may have had an effect on the higher observed Q_{10} values for Days 4 – 6. However, R_{NO3} tended to decrease at lower temperatures with increasing time since resaturation; at temperatures <20 °C, mean R_{NO3} on Days 1 – 6 after rewetting were 14.2, 13.1, 11.5, 10.4, 10.5, and 11.2 g N m^{-3} d^{-1}.

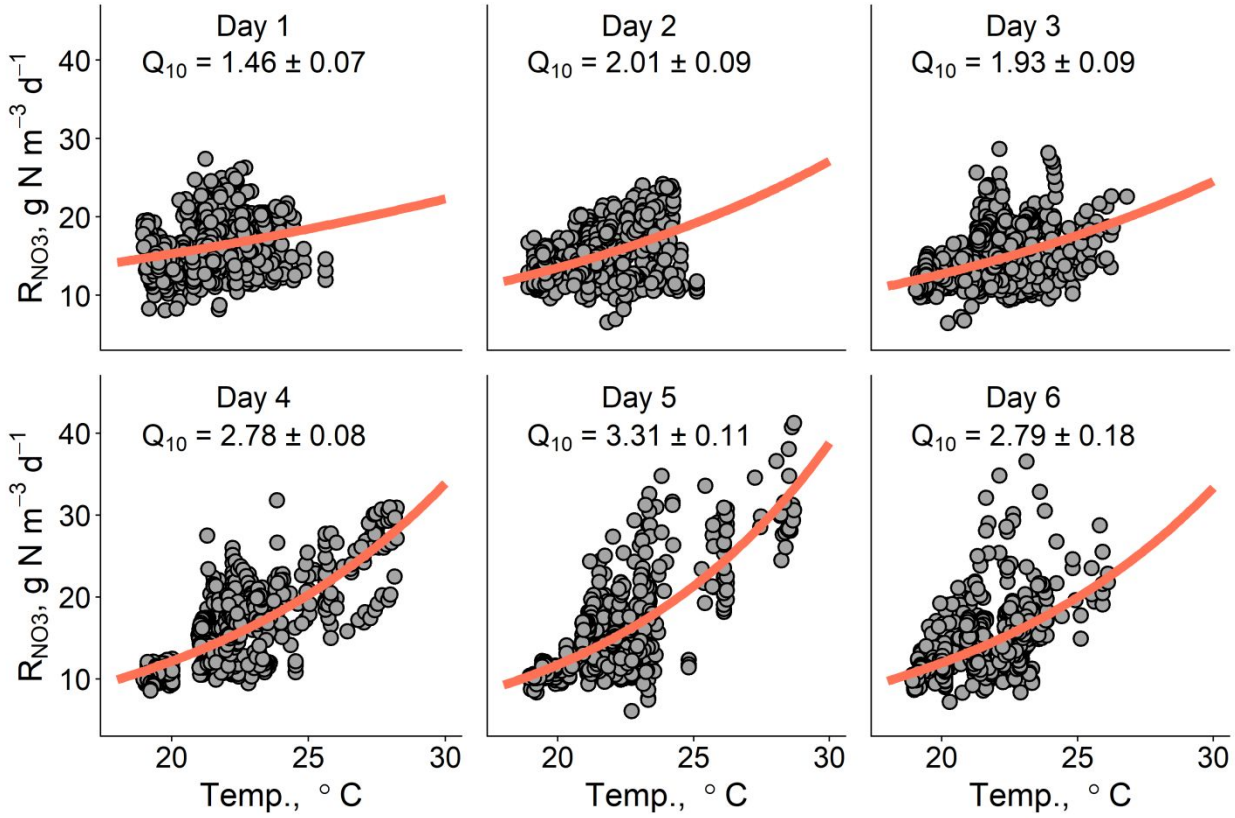


Fig. 6. Relationship of volumetric NO_3^- removal rates, R_{NO3}, with temperature in the NCSU experiment when separating data according to number of days since the 8 h DRW cycle (i.e., the top left panel includes only measured R_{NO3} values within the first 24 h after resaturation of woodchips). Data for each day after rewetting were pooled irrespective of year (2017 and 2018 data combined).

4. Discussion

4.1 Long-term changes in Q_{10}

Data from both experiments support the initial hypothesis that temperature sensitivity of NO_3^- removal in woodchip bioreactors increases over time. The most likely explanation for these observed long-term increases in Q_{10} is changes in carbon quality of the woodchips over time. Ghane et al. showed the relative proportion of lignin in woodchips in a field bioreactor increased over time, with decreasing content of cellulose and hemicellulose [16]. Breakdown of recalcitrant, lignin-heavy organic material through anaerobic respiration has been shown to be negligible. This is possibly due to the inability of the anaerobic pathway to breakdown the complex linkages that occur in lignin [43, 44]. Limited degradation of the woodchips by denitrifiers may be as much due to the carbon structure as its composition, with much of the

cellulose in woody material protected by a lignin "sheath" that is resistant to enzymatic attack [45]. Assuming more bioavailable cellulose and hemicellulose was lost from the woodchips over time in the UPCT and NCSU experiments, denitrifiers were less efficient at metabolizing the remaining carbon to achieve reduction of NO_3^- to gaseous N. Changes in the Q_{10} value were mostly driven by decreased R_{NO3} at lower temperatures, rather than increases in R_{NO3} at higher temperatures, suggesting that denitrification rates at higher temperatures were less affected by changes in carbon quality. Declines in nitrate removal rates in aged woodchips at low temperatures is an important aspect of woodchip bioreactors that should be considered for their use in cold weather climates. For example, woodchips bioreactors have been widely adopted in the Midwest United States as a water quality BMP for NO_3^- load reductions in drainage water. Temperature of tile drainage water in this region, however, is low for most of the year, particularly during the months of April – May (4 – 10 °C) [4] when as much as 40% of annual tile flow can occur [46]. The highest losses in efficiency for woodchip bioreactors over time will occur at the lowest temperatures.

While the increasing Q_{10} values can be considered an indicator of decreasing carbon quality, a separate indicator was the residual model error of the Q_{10} relationship when fitting the relationship in Equation 3. In the UPCT experiment, this residual model error decreased over time from 4.3 g N m^{-3} d^{-1} during Day 30 – 395 to 3.0 g N m^{-3} d^{-1} during Day 365 – 730. A similar trend was observed in the NCSU data from Day 30 – 287 (2017) to Day 480 – 558 (2018), where model error did not change in the SAT group but decreased from 3.8 to 2.6 for the DRW columns. Change in the model error can illustrate temperature sensitivity of NO_3^- removal, as more of the R_{NO3} variability was able to be explained by temperature only when carbon quality was low. A simple temperature-dependent relationship was less capable of explaining R_{NO3} variability when carbon availability was high. Temperature only explained 54 – 85 and 26 – 47% of R_{NO3} variability in the UPCT and NCSU experiments, respectively, indicating there were likely additional factors (e.g., carbon availability) affecting NO_3^- removal rates.

4.2 Effect of drying-rewetting cycles on Q_{10}

Drying-rewetting cycles had both short and long-term effects on Q_{10}. In the NCSU experiment, change in Q_{10} from the Day 30 – 297 to Day 480 – 558 in constantly saturated SAT columns was low (0.25). This contrasted with the larger change in Q_{10} for DRW columns (0.71), as the weekly

aerobic periods would have resulted in greater degradation of and carbon loss from the woodchips. The short-term effect was also apparent, as Q_{10} generally increased with each subsequent day after woodchips were resaturated. This was most likely caused by the gradual flushing or consumption of aerobically-produced DOC following the DRW cycle, consistent with previous findings showing DOC leaching highest immediately following DRW cycles and decreasing quickly (i.e., within days) after resaturation [47 – 49]. Byproducts of incomplete decomposition of organic matter (e.g. DOC) are typically lower molecular weight electron donors [50 – 52], with lower molecular weight organic compounds more bioavailable for certain microbes [53 – 55]. The DRW cycles exposed the lignin-heavy woodchips to aerobic conditions while the media was unsaturated, producing more labile carbon as a result of the more rapid aerobic degradation. Once the media was resaturated and anaerobic conditions resumed, denitrifiers had access to higher quality carbon which led to higher R_{NO3}.

The effect of the DRW cycle was also apparent in the UPCT experiment (Fig. 2). During Days 30 – 395, Q_{10} following the 96 h unsaturated period changed with number of days following resaturation, with the greatest Q_{10} in the third batch run of the week. The same was true during Days 365 – 730, with larger increases in Q_{10} between consecutive batches. Degree of decomposition of the UPCT woodchips during Days 365 – 730, after the fresh woodchips had been used for one year, would be most comparable to the aged NCSU woodchips. There was a large increase in Q_{10} between Batch 1 and Batch 2 during Days 365 – 730 in the UPCT bioreactors (0.55, Fig. 2, hollow triangles), comparable to the increase in Q_{10} from Day 1 to 2 in the NCSU experiment (0.56, Fig. 6). Similarly, the increase in Q_{10} from the second to third day, in both experiments, was 0.11 – 0.12, suggesting the largest changes in carbon quality occurred in the first 24 h following the DRW cycle as aerobically-produced carbon was leached or consumed. Residual model errors fitting the data to Equation 3 also decreased with time since the DRW cycle for both experiments. Using the Q_{10} values from Day 365 – 730 of the UPCT data (Fig. 2, hollow triangles) and the NCSU data (Fig. 6), the relationship of Q_{10} versus number of days since rewetting was well-fitted by a natural log equation of $Q_{10} = 0.62 * \ln(t) + 1.38$ (R^2 =0.95) for UPCT and $Q_{10} = 1.05 * \ln(t) + 1.18$ (R^2 =0.90) for NCSU, where t is number of days since rewetting.

Higher carbon quality and/or availability can explain the observed long-term increases in Q_{10} as woodchips aged (Fig. 1 and 5) and with elapsed time since a DRW cycle (Fig. 2 and 6).

Denitrifiers would have had greater access to more labile carbon when woodchips were less aged (i.e., higher cellulose content) and immediately following unsaturated periods that made lower molecular weight carbon more available via aerobic processes. Once woodchips were resaturated, and anaerobic conditions reestablished, higher denitrification rates would be observed due to the greater carbon availability. This hypothesis attributes differences in carbon availability solely to changes in quality of the woodchip-derived carbon directly accessible to denitrifiers. This differs from the conclusion previously reached by Nordstrom and Herbert [25], which also saw long-term increases in Q_{10} for NO_3^- removal in woodchip bioreactors. The authors concluded changes had occurred in the microbial community composition and/or the degree of cross-feeding between denitrifiers and fermenting bacteria. This was based on the authors' assumption that denitrifiers in woodchip bioreactors rely on the byproducts (e.g. sugars, volatile fatty acids, H_2) of upstream fermenters for electron donors. Although it has been shown that cross-feeding between fermenters and denitrifiers occurs [56], it is possible that there are other mechanisms explaining the increase in temperature sensitivity of denitrification over time. The present study suggests a separate hypothesis, independent of fermentation activity, that accounts for these long-term changes in Q_{10}. A significant portion of fresh woodchips is comprised of cellulose (35 – 56%) [57 – 59], relative cellulose content of woodchips decreases over time (23 – 31% after four years) [16], and, in an oxygen-free environment, a pure culture of denitrifiers is capable of using cellulose as a carbon source [60]. This rationale for the long-term change in NO_3^- removal rates is consistent with the previously established carbon quality-temperature hypothesis, that respiration rates are increasingly sensitive to temperature as carbon quality of the organic matter decreases. This hypothesis also explains the observed short-term changes in Q_{10} immediately following a DRW cycle, since carbon availability would be highest immediately following the unsaturated period in which aerobic processes likely occurred. It is possible that either or both processes (i.e., cross-feeding of fermenters and denitrifiers, short/long-term changes in carbon quality of the media) are occurring in woodchip bioreactors.

Although the present study observed changes in Q_{10} in response to DRW cycles in a high C content substrate (i.e., woodchips), the results are applicable to understanding processes driving organic decomposition in soils. The fact that elapsed time since resaturation of woodchips had an impact on Q_{10} may help explain variability in the literature regarding Q_{10} for respiration of organic matter. A number of studies have indicated that factors other than carbon quality must be

driving changes in Q_{10} [61 – 63]. Peaks in denitrification rates can occur immediately following DRW cycles upon rewetting [49]. Changes in moisture content via DRW cycles, exposure of carbon to aerobic breakdown, and subsequent leaching of soluble organics could explain the variability of Q_{10} in the literature that cannot be explained by carbon quality alone.

4.3 Woodchip degradation and carbon availability

Several factors could cause woodchips to degrade at different rates, and simply using the age of woodchips to predict Q_{10} over time may not be accurate. Moorman et al. showed that shallower woodchips more frequently exposed to aerobic conditions in a field bioreactor had 55% greater carbon loss relative to those in deeper woodchips [28]. In the UPCT experiment, woodchips were exposed to a 96 h unsaturated period once each week. It is possible that woodchips exposed to shorter unsaturated periods would have a lower increase in Q_{10} from the first to second year, relative to the 0.46 increase seen in UPCT bioreactors (Fig. 1). However, despite DRW columns in the NCSU experiment being exposed to a much shorter 8 h DRW cycle, relative to the UPCT bioreactors, they saw a larger increase in Q_{10} (0.71) from the first to second year. This may have been due to the fact that NCSU columns were operated in continuous flow, rather than in batch experiments. Continuously receiving aerated water (~ 8 mg DO L^{-1}) may have caused NCSU woodchips to degrade faster than if they had been operated in 24 h batch experiments. Woodchips in NCSU columns were also left unsaturated between the 2017 and 2018 experiment, and likely experienced greater rates of degradation over this period. A third factor that may have increased the rate of degradation of the UPCT woodchips was the use of saline brine in which sodium concentrations ranged from 2,600 – 5,000 mg Na L^{-1}. Previous research has shown that high salinity [64] or sodicity [65] can increase the breakdown of organic matter. Indeed, previous experiments at the UPCT facility showed DOC in the effluent of woodchips was greater as brine became more concentrated. Changes in temperature sensitivity over time would be site specific and depend on various factors, including degree of exposure to aerobic conditions and water chemistry.

In both experiments, rates of DOC release increased at higher temperatures. Values of Q_{10} for DOC release during Days 30 – 395 and 365 – 730 of the UPCT experiment were 1.75 and 1.52, respectively (Supplemental Fig. S1); Q_{10} of DOC release for NCSU SAT and DRW groups were 3.44 and 3.42 during Days 30 – 287 and 2.84 and 2.65 during Days 480 – 558 (Supplemental

585 Fig. S3). While part of the temperature response of R_{NO3} would have been related to the
586 efficiency of denitrifiers to metabolize carbon, the effect of temperature may also have been
587 confounded with higher aerobic decomposition rates when woodchips were unsaturated resulting
588 in greater carbon availability, linking the temperature sensitivity of aerobic and anaerobic
589 respiration. Q_{10} values for denitrification in woodchip bioreactors combine the effect of several
590 processes which are also affected by temperature, such as those which increase carbon
591 availability of woodchip-derived carbon (i.e., aerobic breakdown during unsaturated conditions).

592 Increased C availability at higher temperatures due to aerobic breakdown may explain dynamic
593 trends in Q_{10} during the UPCT experiment. The overall Q_{10} and Q_{10} for each batch run increased
594 until Day ~140 – 150 at which point Q_{10} values reached a plateau (Fig. 3). Subsequently, Q_{10} for
595 Batch 1 began to decrease after Day ~210, while Q_{10} increased for Batch 2 roughly 20 days later
596 as temperatures were increasing during the summer months. This could be explained by greater
597 carbon availability via more efficient aerobic breakdown at warmer temperatures, with
598 denitrifiers able to consume nearly all of the aerobically-produced carbon during Batch 1 and
599 leaving less available for the subsequent Batch 2. The fact that most of the increase in Q_{10} for all
600 batches occurred during the first ~150 days is consistent with previous findings that most of the
601 declines in NO_3^- removal in woodchip bioreactors occurs relatively rapidly (<1 year) and is
602 relatively stable after this initial leaching period of more readily consumed carbon (i.e., cellulose
603 and hemicellulose). It is also possible that changes in the microbial community during Days 150
604 – 230 that caused Q_{10} changes in Batch 1 and 2.

605 **4.5 Temperature dependence of Q_{10}**

606 Several studies have reported higher [5] or lower [25] Q_{10} values at higher temperatures, and that
607 Q_{10} can depend on magnitude of or total range in temperature observed [66]. During Days 365 –
608 730 (second year), at minimum temperature values of 10 to 11 °C (x-axis), calculated Q_{10}
609 generally increased with increasing range in temperature values (y-axis), indicating data
610 collected at low temperatures over a small range in temperature may bias Q_{10} values towards
611 underestimation. This is possibly due to the fact that variability in observed rates (as affected by
612 measurement uncertainty or experimental variability) are larger relative to the total temperature-
613 induced change in rates, when temperature range is small. This was seen in the higher
614 uncertainty of the Q_{10} values at smaller ranges of the temperature interval (Supplemental Fig. S2,

y-axis), which may have explained the higher variability in Q_{10} values at different values of minimum temperature when the range in temperature of the interval was only 5 – 6 °C (bottom two rows of tile plots in Fig. 4). In both years of the UPCT experiment, uncertainty of Q_{10} was <5% when the range in the temperature interval was ≥10 °C. Researchers calculating Q_{10} values when range in temperature is small should consider this additional uncertainty when drawing conclusions.

Using the results of this study as an example, temperature range during Day 480 – 558 of the NCSU experiment (20.5 – 24.7 °C) was smaller than during Day 30 – 287 (18.6 – 29.0 °C). Recalculating Q_{10} during Day 30 – 287, subsetting the data to the same temperature interval seen during Day 480 – 558, Q_{10} values were 1.41 and 1.27 for SAT and DRW groups, respectively, indicating that the change in Q_{10} from the first to second year may have been much greater than initially thought (Fig. 5). Additionally, in NCSU bioreactors there was a lower total range in temperature seen in Days 1 – 3 after rewetting (19 – 25 °C, Fig. 6), relative to Days 4 – 6 after rewetting (19 – 29 °C). Recalculating Q_{10} values by subsetting the data to the smaller temperature range (19 – 25 °C), Q_{10} for Days 1 – 6 were 1.53, 2.11, 2.07, 3.37, 3.83, and 2.79, respectively, which still showed an increase in Q_{10} with number of days since resaturation of the woodchips.

5. Conclusions and management considerations

Temperature sensitivity of NO_3^- removal rates in woodchips bioreactors increased as woodchip aged in both experiments, showing that woodchip age is an important parameter in understanding the effect of temperature on NO_3^- removal and when calculating Q_{10}. Similarly, DRW cycles caused brief increases in NO_3^- removal that tended to decrease temperature sensitivity immediately after rewetting, which was modulated by time elapsed since the DRW event, as shown by higher Q_{10} values as time since resaturation increased. Both trends can be attributed to decreasing bioavailability of carbon for anaerobic denitrification and are consistent with the carbon quality-temperature hypothesis. Soluble organic carbon in the effluent also increased at higher temperatures, particularly after DRW cycles, which was coincidental with increases in NO_3^- removal rates. This finding suggested that microbial activity was stimulated at higher temperature during unsaturated conditions, leading to a surplus of low-molecular weight soluble organic carbon compounds via incomplete respiration, which, in turn, may have played some

part in the temperature response of denitrification during the subsequent flooding phase. Although it is clear that DRW cycles produce increased nitrate removal rates, the management method is likely to lead to more rapid degradation of the media. Implementing DRW cycles may also require additional resources (e.g., equipment, labor) to regularly drain and resaturate media. Water quality managers would need to consider these factors when choosing between a continuously saturated system or one with intermittent DRW cycles.

Short and long-term changes in temperature sensitivity in woodchip bioreactors should be considered both in the context of agricultural water management and its behavior under changing climactic conditions. Water quality planners should consider declines in NO_3^- removal efficiency over time will be greatest at lower temperatures. Similarly, depending on regional impacts of climate change, more prolonged dry periods would lead to greater degradation under unsaturated conditions of woodchips since field woodchip bioreactors are often located above the water table and drainage lines.

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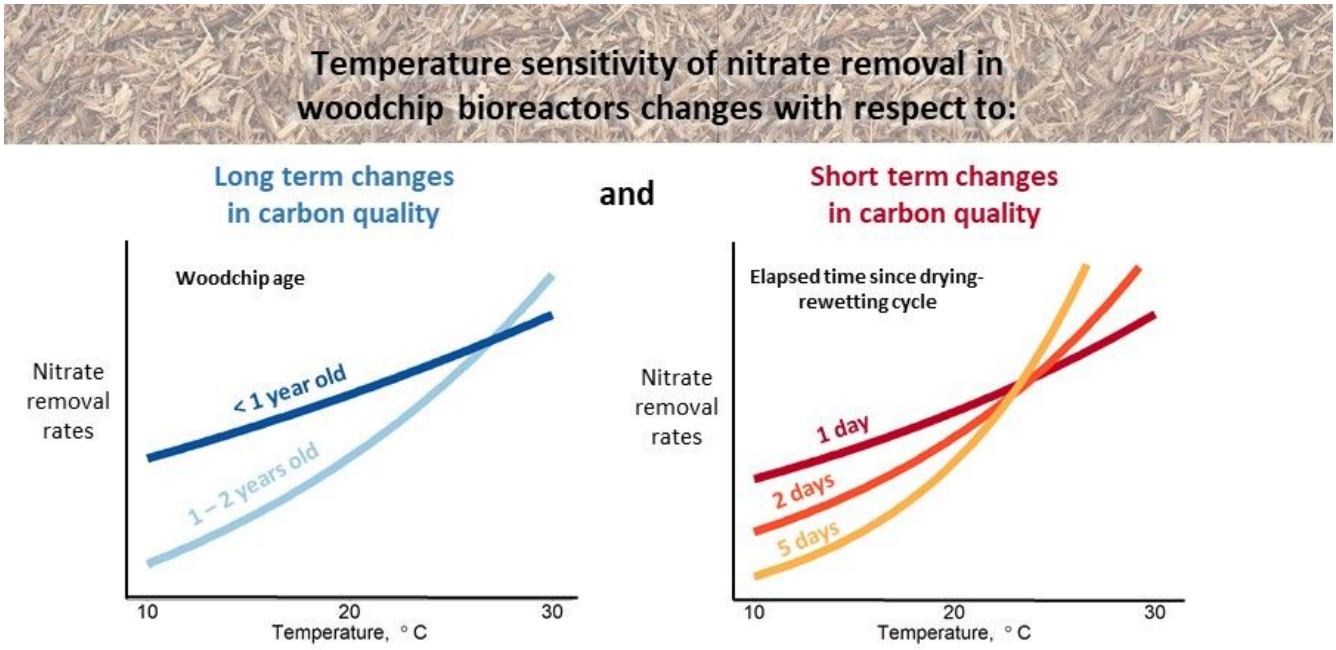

Description of novelty of work for table of contents : Temperature sensitivity of nitrate removal in woodchip bioreactors changes according to short and long-term changes in carbon quality.