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Growth and Uptake Dynamics of Selenastrum Capricornutum Parameterized by Percent Nitrogen

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ABSTRACT

Growth and Uptake Dynamics of *Selenastrum Capricornutum*

Parameterized by Percent Nitrogen

by

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Utah State University, 1977

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Batch cultures of *Selenastrum capricornutum*, PRINTZ were perturbed by dilution and nutrient spiking routines to obtain a wide variation of introcellular nitrogen levels (2 to 7 percent) under dynamic conditions. The relationship between specific growth rate (dry weight) and cellular nitrogen content (percent nitrogen) was investigated by regression analysis and continuous mathematical simulation.

Linear regression analysis resulted in good correlation ($r^2 = 0.817$) between cellular percent nitrogen and specific growth rate. Continuous simulation revealed the relationship was least accurate for the lag phase of growth. The observed deviations always occurred when the cellular nitrogen content was increasing. This indicated that the rate of change of cellular percent nitrogen, as well as it's absolute level, affects the observed growth rate. Data points were separated into two groups having decreasing and increasing rates of change in cellular nitrogen content respectively. Separate regression analyses were performed on each data set. A strong correlation ($r^2 = 0.883$) between specific growth rates and percent nitrogen was obtained for the first set of data. The second set of data exhibited a constant low specific growth rate ($\mu = 0.05$

per day) for cultures with low percent nitrogen content. At higher nitro-
gen contents, the relationship between growth and nitrogen content was
identical to the results in the first data set. The transition zone
between reduced and normal growth occurred in a range of 3 to 4 percent
nitrogen content.

(99 pages)

INTRODUCTION

Algal models are being applied to answer increasingly sophisticated questions about eutrophication. Species dominance, as well as, biomass is a significant factor in defining the effect of algae upon the recreational and consumptive values of our streams and reservoirs. Models capable of giving results of a high resolution must be developed to answer questions concerning species dominance. Further, application of models to dynamic river and estuary systems requires algal models that realistically represent kinetics of growth and uptake under rapidly changing conditions.

First order and Monod type relationships are commonly being utilized to represent algal kinetics in a number of water quality models (Grenney, Porcella, and Cleave, 1976). In most cases the basic assumption of constant yield is maintained. This assumption is not compatible with observed phenomena. The widely recognized phenomenon of luxury uptake (Gerloff and Skoog, 1954; Fogg, 1959; Daley and Brown, 1973; Droop, 1973; Eppley and Renger, 1974; Malone and Garside, 1975; Reynolds et al., 1975), variations in cell composition (Gerloff and Skoog, 1957; Caperon, 1968; Thomas and Dodson, 1972; Daley and Brown, 1973; Droop, 1973; Eppley and Renger, 1974) and growth in a nutrient depleted medium (Gerloff and Skoog, 1954; Fogg, 1959; Rhee, 1973; Daley and Brown, 1973; Eppley and Renger, 1974) cannot be represented by models assuming a constant yield.

Observations of luxury uptake, variations in cell composition, and growth in nutrient depleted media have prompted a number of authors (Caperon, 1968; Thomas and Dodson, 1972; Grenney et al., 1973; Droop,

1973) to suggest conceptual models which include internal storage pools of nutrients. A lack of understanding of the precise composition of the proposed storage pools have hindered development of such models.

This paper investigates the feasibility of utilizing variations in nitrogen content of cells to represent storage phenomena. More specifically, the relationships between growth and uptake dynamics and percent nitrogen in batch cultures of *Selenastrum capricornutum*, PRINTZ are examined. The objective here is to determine the potential of percent nitrogen as a basis for a realistic algal model capable of dealing with dynamic situations and questions of species competition to a high degree of resolution.

REPRESENTATION OF STORAGE PHENOMENON BY

PERCENT NITROGEN

The approach of utilizing variations in cell nitrogen content to represent phenomena of storage as related to the kinetics of growth and uptake, gain support from the data presently available on the patterns of nitrogen storage. Fogg (1959) reviewed the metabolic patterns associated with nitrogen in algae. Noting decreases in inorganic nitrogen, free amino acids, soluble peptides and chlorophyll during periods of nitrogen limitation, he suggested that nucleic acids and proteins were maintained at the expense of less essential nitrogenous compounds. This definition of the storage pool is compatible with recent work which has confirmed storage of inorganic nitrogen (Malone and Garside, 1975) and variations of nitrogenous pigments (Bunt, 1969; Eppley and Renger, 1974; Vasconcelos and Fay, 1974) with nitrogen availability. This is also compatible with the theory of Droop (1973) that nitrogen storage occurs in organic forms. However, Fogg (1959) concluded there was little evidence to show whether decreases in nitrogen content were reflected equally in all nitrogen fractions or whether some substances were affected more than others. This uncertainty was more currently emphasized by Caperon (1968, p. 871):

> The hypothesized internal reservoir is not necessarily a physically separate identity any more than a diffuse component of the protoplasm nor is it implied that the internal supply is in the form of nitrate ions.

We may, therefore, reasonably assume that the storage phenomena involve both inorganic and organic nitrogenous compounds, and that the

proposed internal storage pool exists 1) in the ability of the algal culture to shift the distribution of internal nitrogen such that essential forms dominate and/or 2) in their ability to function through variations in levels of essential forms.

Two approaches for representing the storage phenomenon were considered. The first was the development of an algal model consisting of a number of internal subcompartments (Figure 1). This approach was outlined by Grenney et al. (1973). This model defined three internal subcompartments, inorganic nitrogen (N_1), organic nitrogenous intermediates (N_2), and protein (N_3). The size of the population was measured by the concentration of protein in the environment. The external concentrations of nitrate and excreted nitrogenous organics were identified as N_I and N_o, respectively. The coefficients G_I, G_1, G_2, K_3, K_4, K_5, and K_6 represented reaction rates among subcompartments.

Such an approach is compatible with the metabolic patterns associated with nitrogen storage and assimilation. In mathematical terms it has the flexibility to represent many of the responses of phytoplankton under dynamic conditions. However, the practical difficulties of defining and measuring internal components for verification of internal reaction rates discourages use of such a vigorous approach. The use of protein to measure phytoplankton concentrations makes conversion to biomass (measured as dry weight) difficult because the protein content of algal cells has been observed to vary (Fogg, 1959).

The second approach was the use of nitrogen content to represent storage phenomena. Shiroyma, Miller and Greene (1975) have observed a linear relationship between maximum yield and nitrogen concentration
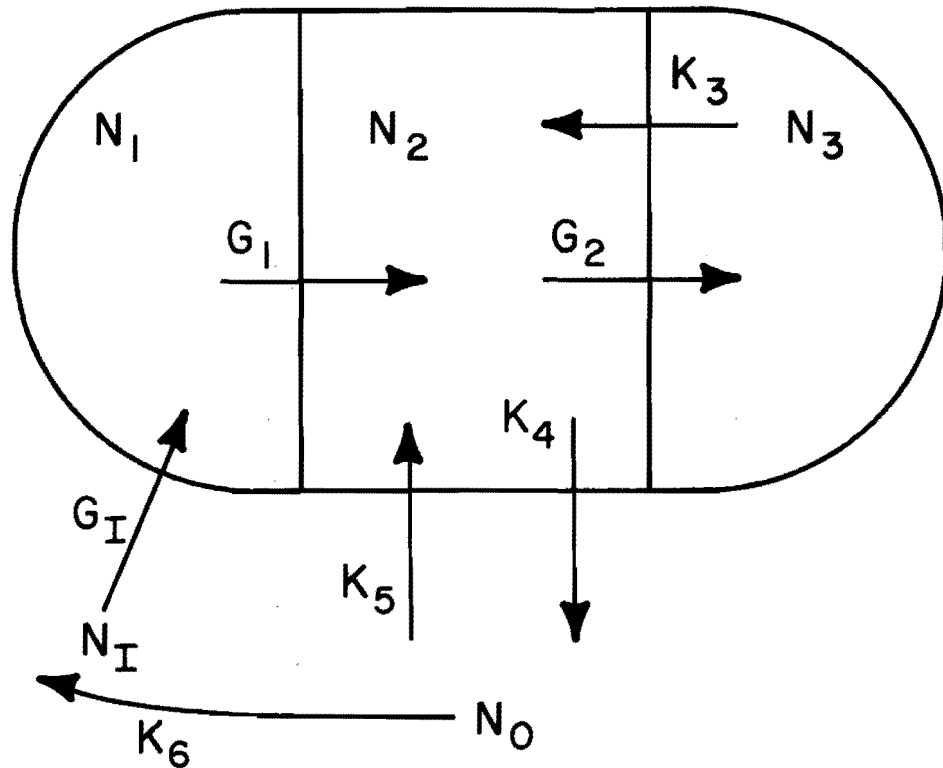
Figure 1. Schematic diagram of a subcompartment model of a phytoplankton population. (Proposed by Grenney et al., 1973.)

under controlled laboratory conditions. They concluded that 0.001 mg-N/1 of total soluble inorganic nitrogen will produce a maximum yield of 0.038 mg/1 dry weight of the alga *Selenastrum capricornutum* under constant laboratory conditions. From this relationship the cell nitrogen content of the cultures can be calculated to be 2.6 percent as the growth of the culture approaches zero.

The works of Gerloff (Gerloff and Skoog, 1954, 1957; Gerloff and Krombholz, 1966) demonstrate a relationship between percent nitrogen and exponential growth. In his efforts to utilize nutrient content to pre-dict nutrient limitations, he defined levels of internal nitrogen and

phosphorus that were necessary to permit optimum growth of the blue green alga, *Microcystis aeruginosa*. He established that a 4.0 percent nitrogen content was necessary for exponential growth to occur in cultures of *M. aeruginosa*.

The use of nitrogen content to represent the storage phenomenon requires not only that the potential for growth be measurable but also that the potential can be accurately related to kinetics of growth and uptake of the phytoplankton. Utilizing the subcompartment model (Figure 1) for purposes of illustration, it can be seen that an infinite number of combinations of internal nitrogenous components can be envisioned for a given percent nitrogen value. This argument indicates that percent nitrogen would be a poor indicator of physiological condition since the internal distribution would vary significantly with the nutrient availability in the history of the cell.

Fortunately, it is reasonable to assume that internal nitrogenous forms are synthesized and depleted in an established order of priority. Assuming the internal reaction rates are sufficiently fast, a given percent nitrogen value would result in a consistent internal distribution of nitrogenous compounds; the nitrogen content of a culture would reflect a certain physiological state which in turn could be related to uptake and growth kinetics. Caperon and Renger (1974) have illustrated a hyperbolic relationship between nitrogen to carbon ratios (N/C) and specific growth rate for continuous cultures. Since percent nitrogen is closely related to the N/C ratios, a similar relationship will hold for percent nitrogen and growth rates under continuous culture conditions.

The works cited in the above discussion indicate the potential of cell nutrient content as a basis for predicting alga kinetics. A relationship between maximum and minimum growth and cell nitrogen content under batch culture conditions has been previously demonstrated. This information supports the hypothesis that a relationship between growth rates and nitrogen content similar to that found for continuous cultures exists for more dynamic batch culture conditions.

PERCENT NITROGEN MODEL

Figure 2 illustrates the conceptual percent nitrogen model. The diagrams illustrate a theoretical unit cell with two internal nitrogen components. The component labeled "NMIN" represents the minimum nitrogen fraction found when the growth rate is zero due to nitrogen limitation. The component "N_s" is the nitrogen fraction in excess of NMIN. This component represents nitrogen reserves which reflect additional potential for growth.

This conceptual model does not represent precisely the nitrogen storage phenomenon previously described. In contrast to the subcompartment model (Grenney et al., 1973), the percent nitrogen model lumps all internal nitrogen components into two classifications, NMIN or N_s. Questions concerning exact internal components or mechanisms need not be answered, nor is exact mathematical representation of the numerous mechanisms required. This permits the development of an uncomplicated set of differential equations based on information presently available.

The following set of differential equations can be utilized to describe the kinetics of uptake and growth related to the percent nitrogen model:

$$\frac{dx}{dt} = U*N_s*X \quad\quad\quad\quad\quad\quad\quad (1)$$

$$\frac{dN}{dt} = -G*N*X = -\frac{dN_c}{dt} \quad\quad\quad\quad (2)$$

$$N_s = (\%N-NMIN) \quad\quad\quad\quad\quad\quad (3)$$

in which

U = growth rate constant (days^{-1})

N_s = excess intracellular nitrogen

NITROGEN RESERVES
MAINTAINED

EXTERNAL SOURCES AND
INTERNAL RESERVES DEPLETED
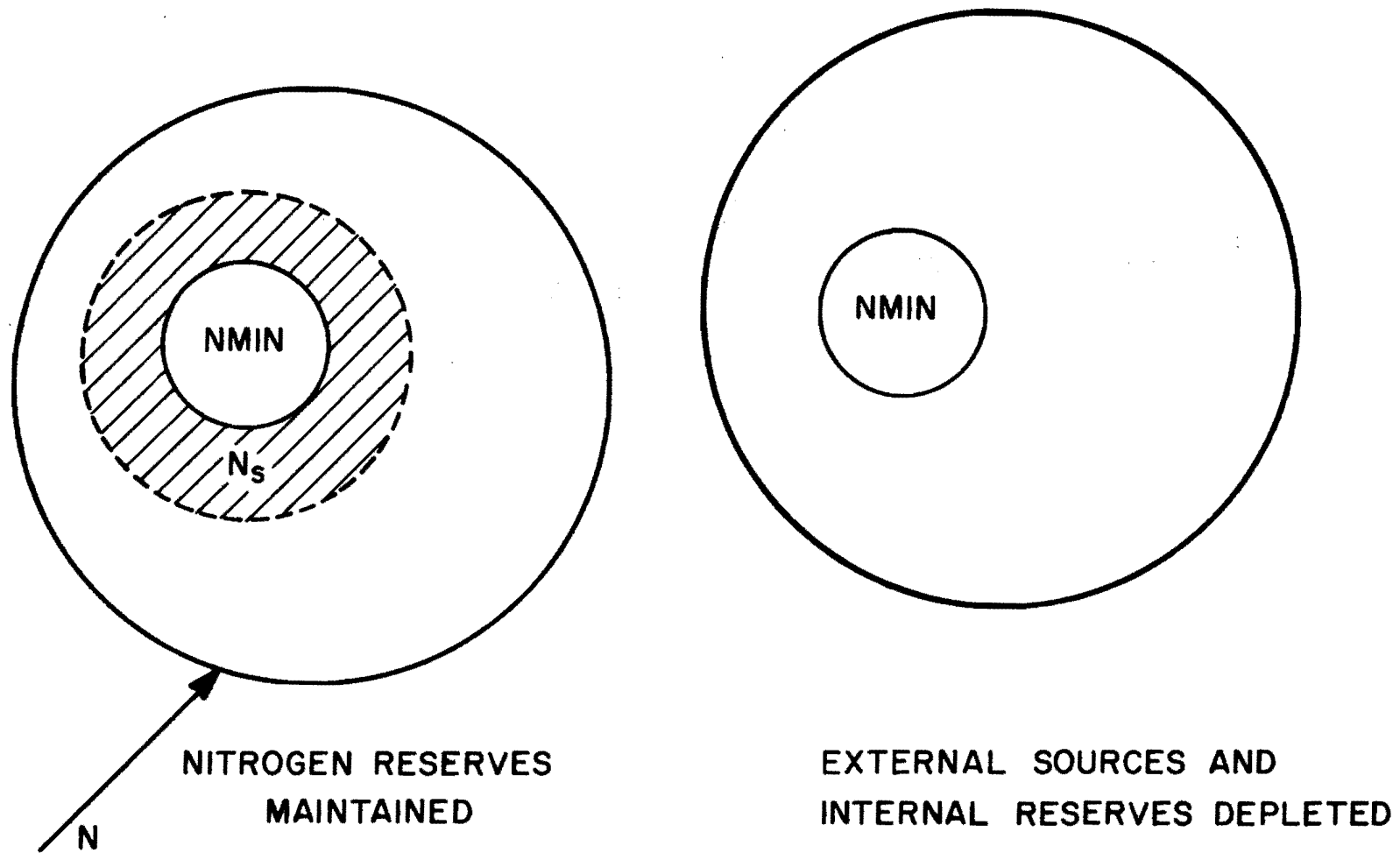
Figure 2.   Conceptual percent nitrogen model.

X = algal biomass (mg/1 dry wt.)

G = uptake rate constant (liters/mg-day)

N = extracellular inorganic nitrogen (mg-N/1)

$\%N$ = percent nitrogen of algal biomass

$NMIN$ = minimum percent nitrogen

N_c = intracellular nitrogen (mg-N/1)

Equation 1 implies that the specific growth rate (EPA, 1971) is directly proportional to the excess intracellular nitrogen level, N_s. This linear relationship differs from the hyperbolic relationship utilized by Caperon and Renger (1974) to describe the relationship between N/C ratio's and specific growth rate. There are a couple of justifications for this deviation. First, despite the loss of one degree of freedom, the linear relationship appeared to fit the data of Caperon and Renger (1974) as well as the hyperbolic. Although a hyperbolic relationship may be assumed to apply throughout a full range of nitrogen availability, linear characteristics would be exhibited in the range of severe nitrogen limitation.

Equation 2 implies that the uptake rate of external nitrogen depends upon the external nitrate level and algal biomass of the system. Droop (1973) and Rhee (1973) have suggested that uptake rates vary as a function of both internal and external nitrogen levels. There is no doubt in the mind of the author that at some point the uptake rate will be affected by the internal nitrogen levels. However, observations made during the course of this study have not indicated that there is any relationship. The point at which internal nitrogen stores begin inhibit uptake activities must lie beyond the levels obtained in nitrogen limited cultures.

Equation 3 defines the storage component, N_s, to be the difference between the existing cellular percent nitrogen and the minimum level.

Application of Equations 1 through 3 requires the definition of three coefficients; the growth rate constant, U, uptake rate constant, G, and the minimum nitrogen level, NMIN. NMIN can be directly calculated from the maximum yield of the algal cultures under a given set of conditions.

Then U and G can be found by calibration of the model to observed data or through correlation studies.

EXPERIMENTAL METHODS

Unialgal cultures of *S. capricornutum* PRINTZ (from U.S. EPA,
CORVALLIS, OREGON) were maintained in three separate 3-liter low profile
culture flasks at $25 \pm 1^{\circ}C$ and 350 ± 35 foot candles. Flasks were con-
tinually mixed by magnetic stirrers.

The AAM medium (EPA, 1971) increased in strength by a factor of
3.33 was utilized for all experiments. The nitrate was reduced to 10
percent of the level normally found in a triple strength media. This
nitrate level has been shown (Malone et al., 1975) to produce a nitrogen
limited environment. The sodium bicarbonate ($NaHCO_3$) level of the medium
was increased to 84 mg/l as $NaHCO_3$ to aid in stabilization of pH.

The pH was maintained by aeration of the cultures with an air-CO_2
mixture. Carbon dioxide from a gas cylinder (95 percent CO_2) was mixed
with ambient air in a mixing chamber. The ambient air was pumped at a
constant rate. The rate at which carbon dioxide was bled into the mixing
chamber was varied until the target pH of 7.1 was obtained in a control
flask containing algal free medium and pH sensitive dye. The pH of the
medium was defined by a function of the ratio of bicarbonate to carbon
dioxide found in the aerated media. The bicarbonate level was selected
such that the resulting carbon dioxide level at pH 7.1 would be suffi-
ciently high to prevent pH variations due to algal activities. This is
possible since the transfer rate of carbon dioxide across the air-water
interface varies as a function of the difference between existing and
saturated carbon dioxide concentrations. This difference, the carbon
dioxide deficit, results from algal uptake of carbon dioxide. The

maximum carbon dioxide deficit for a culture is defined by either the virtual elimination of carbon dioxide or by a balancing of the transfer and uptake rates. The bicarbonate level selected led to a high carbon dioxide saturation level at pH 7.1. This saturated level was sufficiently high so that the maximum carbon dioxide deficit found in our laboratory cultures was not a large enough fraction of the saturated level to affect the pH determining ratio. The air-CO_2 mixture could therefore remain fixed for the duration of an experiment.

Prior to entering the culture flasks, the air-carbon dioxide mixture was passed through a series of flasks to assure removal of ammonia. The flasks contained, in order; 1.0 N H_2SO_4, distilled water, a weak bi-carbonate solution, and packed glass wool. The stripping system also eliminated significant evaporation from the culture flasks, by water saturation of the air. The air-carbon dioxide mixture was continuously bubbled into the culture flasks. Plugs on the flasks were vented permitting the release of pressure, but maintaining the enriched carbon dioxide levels within the flasks.

Temperature (^{o}C), pH biomass (mg/l dry wt $103^{o}C$), nitrogen content of biomass (percent nitrogen), cell counts (cells/ml), cell volume (cubic microns), optical density (1 inch cell at 750 nm) were measured on samples removed periodically from the three culture flasks. Percent nitrogen was measured on a Coleman (micro-Dumas) nitrogen analyzer. Cell counts and cell volumes were measured by a Coulter counter. Optical densities were read on a Bausch and Lomb Spectronic 20. pH measurements were made with a Corning Scientific Instruments, Model 7, pH meter.

During the course of the experiments, cultures at various stages of growth were diluted with fresh medium to permit examination of specific phases of growth and uptake dynamics. Dilution at intervals specified by experimental design was accomplished by mixing 1 liter of culture with 1 liter of fresh medium. The temperature of the fresh medium was equalized before mixing. Thus the volume of culture in the experimental flasks varied from a maximum of 2.5 liters to a minimum of about 1.2 liters due to sample removal during the course of the experiment.

RESULTS

Two experiments were conducted to provide data for calibration of the algal model. Experiment 1 had a duration of approximately six days. The batch cultures were monitored until 4.7 days, at which time the culture was mixed with an equal amount of fresh medium. The response of the cultures to this dilution was measured until 5.81 days. Table 1 presents the results of Experiment 1 in a summarized form. The dry weight, percent nitrogen, and nitrate values represent the mean values of the three culture flasks. A complete data listing is given in Appendix A.

Difficulty was encountered with pH control during the early part of Experiment 1. The pH varied due to variations in the air-carbon dioxide mixture which resulted from a regulator failure. Addition of a new regulator midway through the experiment corrected the difficulty. The pH varied from 6.5 to 10.0 prior to day 3.55 after which it was stabilized at 7.3 ± 0.1.

Experiment 2 had a duration of 19 days. The cultures were diluted three times during the course of the experiment. Dilutions in Experiment 2 were made during different phases of growth. The first dilution (11.06 days) was made after growth had essentially ceased due to nitrogen depletion. The second dilution (15.67 days) was made immediately after the exponential growth phase. And the third dilution (16.85 days) was made late in the exponential growth phase. Table 2 summarizes the mean values of dry weight, percent nitrogen and nitrate for the three cultures of Experiment 2. The pH varied within the range of 6.9 to 7.3 during the

Table 1. Mean biomass, nitrate, nitrogen content, and standard deviations
for Experiment 1.

Time (days)	Dry Weight (mg/ℓ)	Standard Deviation (mg/ℓ)	Percent Nitrogen	Standard Deviation (percent)	Nitrate[3] (mg/ℓ)
1.83	32.23	2.96	5.07	0.22	0.27
2.10	36.54	2.56	3.86	0.16	0.19
2.67	51.20	0.87	2.54	0.27	0.30
2.85	51.60	1.14	2.79	0.19	0.16
3.16	57.67	1.79	2.51	0.13	0.15
3.55	61.40	0.80	2.61	0.10	0.0
4.18	71.60	2.58	2.23	0.02	0.0
4.70	72.30	1.30	2.43	0.38	0.0
4.73[2]	36.40	0.98	2.09	0.13	0.84
4.88	36.60	1.02	2.79	0.31	0.71
5.05	37.07	1.71	4.05	0.19	0.23
5.20	41.00	0.33	3.83	0.13	0.16
5.52	49.00	2.94	3.35	0.0	0.09
5.81	55.20	1.10	3.13	0.07	0.0

[1]Mean values of 3 separate cultures

[2]Immediately after 1:1 dilution

[3]Calculated value from initial mass of N minus mass of particulate N

Table 2. Mean biomass, nitrate, and nitrogen contents and standard deviations for Experiment 2.

Time (days)	Dry Weight (mg/ℓ)	Standard Deviation (mg/ℓ)	Percent Nitrogen	Standard Deviation (percent)	Nitrate[3] (mg/ℓ)
0.93	3.13	0.24	6.07	0.15	1.38
1.50	9.77	0.71	7.06	0.54	0.88
2.00	23.40	2.20	5.39	0.19	0.31
3.05	47.33	2.56	3.32	0.14	0.0
4.03	58.20	0.87	2.68	0.08	0.0
5.01	69.27	1.32	2.27	0.13	0.0
11.04	79.00	1.58	2.13	0.06	0.0
11.06[2]	38.93	0.51	2.31	0.21	0.83
11.52	38.60	0.58	3.26	0.46	0.47
14.97	77.67	0.62	2.20	0.20	0.02
15.64	80.00	0.56	2.16	0.07	0.00
15.67[2]	38.73	0.81	2.04	0.10	0.80
16.05	40.93	0.65	3.30	0.08	0.24
16.56	55.47	1.05	2.79	0.04	0.04
16.83	59.40	0.71	2.58	0.12	0.06
16.85[2]	30.23	0.21	2.71	0.10	0.80
17.06	30.77	0.80	4.65	0.30	0.19
17.53	44.27	2.61	3.41	0.09	0.11
18.09	55.07	4.24	2.80	0.12	0.08
19.23	66.13	2.31	2.25	0.00	0.13

[1]Mean values of 3 separate cultures

[2]Immediately after 1:1 dilution

[3]Calculated value from initial mass of N minus mass of particulate N

course of Experiment 2. One minor adjustment of the air-carbon dioxide

mixture was made at day 1.50. A complete listing for the data generated

during the course of this experiment is presented in Appendix A.

CALIBRATION OF PERCENT NITROGEN MODEL

The calibration process consisted of empirically defining the three unknown constants contained in Equations 1 through 3: the uptake rate constant (G), the growth rate constant (U), and the minimum percent nitrogen (NMIN).

Correlation studies were conducted between a wide variety of parameters in an attempt to define the unknown coefficients. These correlation studies also shed some light upon the appropriateness of the selected equation forms. Figure 3 illustrates a plot of specific growth rate and percent nitrogen data taken from Experiment 1 after dilution (4.73 – 5.81 days) and Experiment 2. Data points from Experiment 1 prior to dilution (1.83 – 4.70 days) were omitted from this analysis because of the unknown effect of pH variations upon the cultures. The specific growth rate (μ) was calculated from Equation 4:

$$\mu = \frac{\ln (x_2/x_1)}{T_1-T_2} \quad \cdot \quad \cdot \quad \cdot \quad \cdot \quad \cdot \quad \cdot \quad \cdot \quad \cdot \quad \cdot \quad \cdot \quad \cdot \quad \cdot \quad (4)$$

The calculated specific growth rate was then correlated with the average percent nitrogen exhibited by the cultures over the time interval. Individual observations (Appendix A) were utilized for this plot. Linear regression by least squares reveals a correlation ($r^2 = 0.817$) between specific growth rate and mean percent nitrogen. From this correlation it is evident that U = 0.407 and NMIN = 2.31 percent for Equation 1.

No correlation between the uptake constant (G) and percent nitrogen exists in the regime of nitrogen limitation utilized in this experiment (Figure 4). The values of G for this correlation plot were developed from Equation 5:
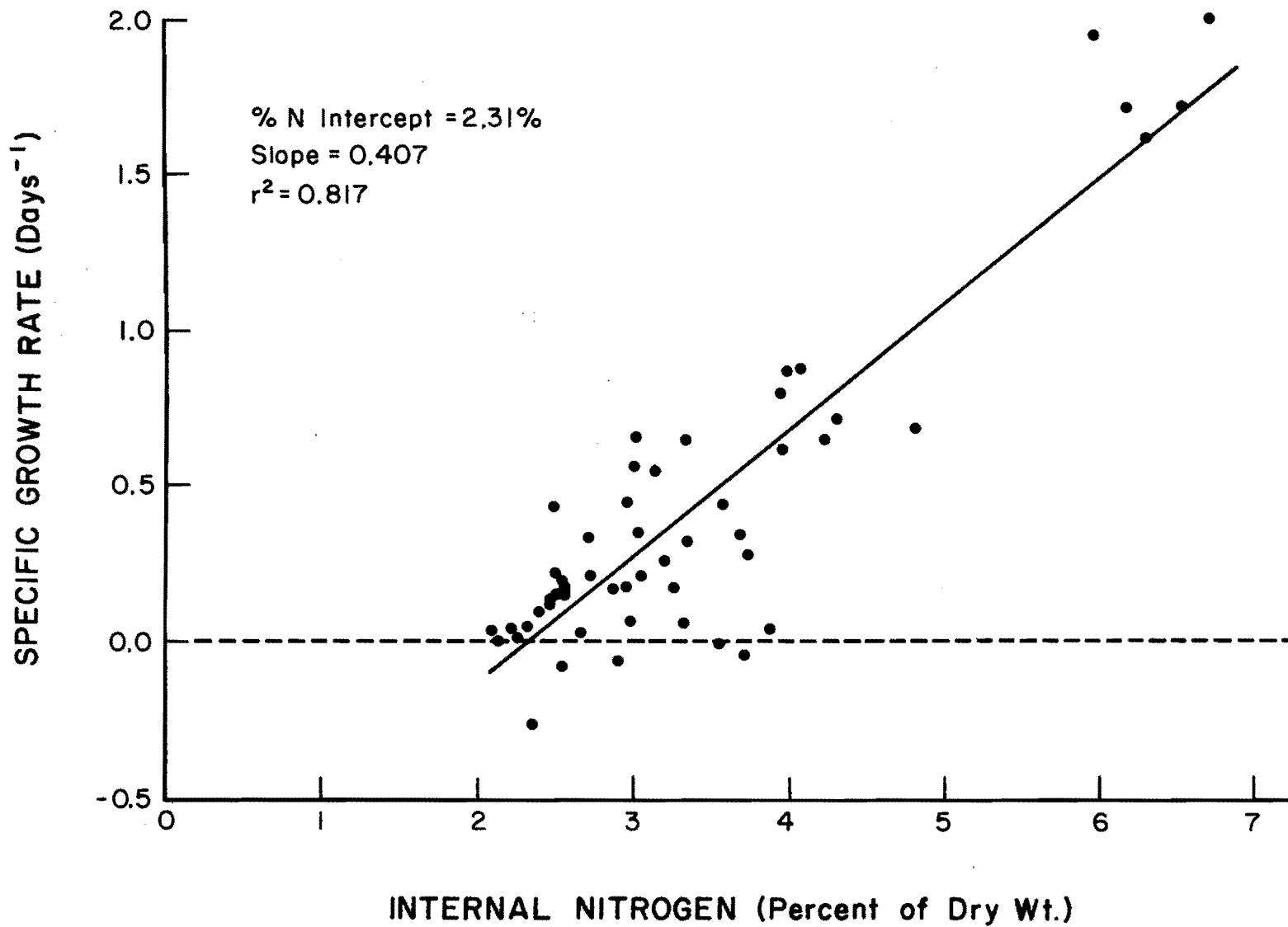
Figure 3. Correlation of specific growth rate to mean percent nitrogen for all data.
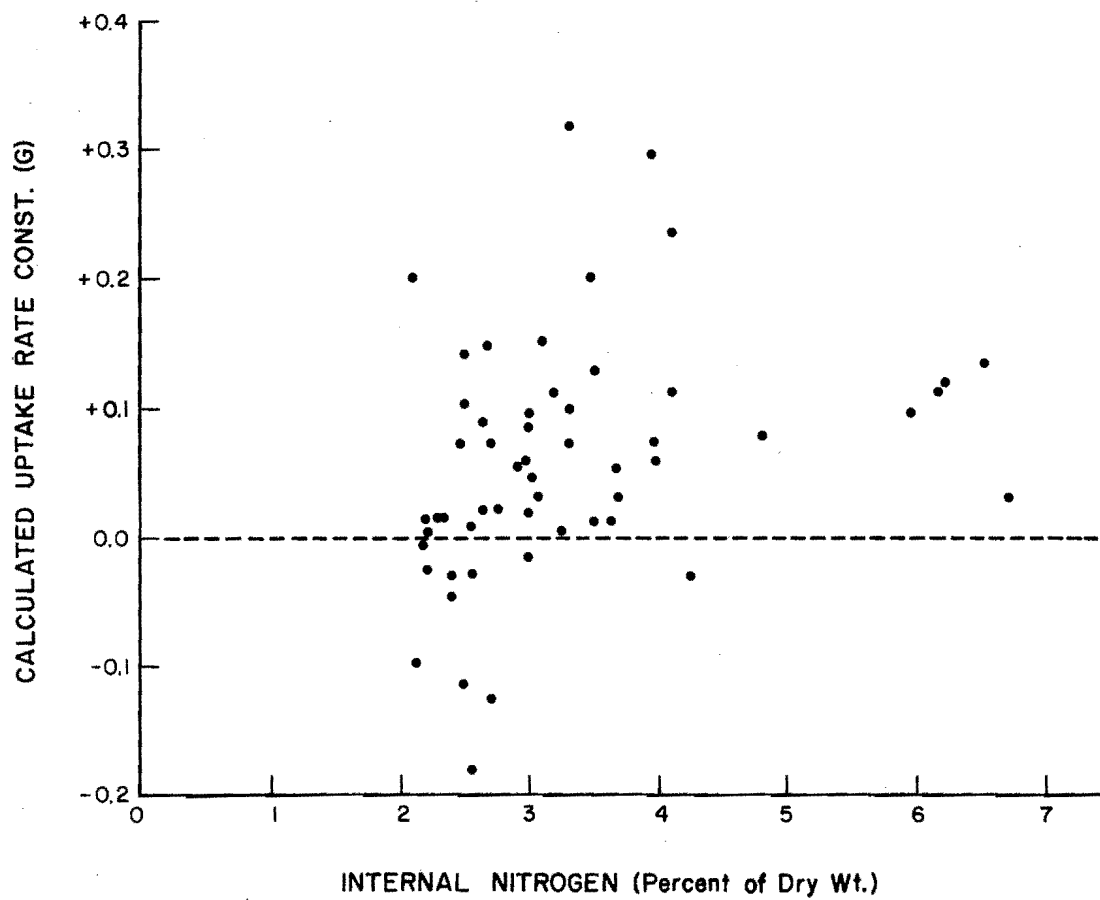
Figure 4. Correlation of uptake rate constant to mean percent nitrogen.

$$G = \frac{\Delta N}{\overline{N} * \overline{X} * \Delta T} \quad \cdots \cdots \cdots \cdots \cdots \cdots \cdots (5)$$

in which $\overline{N}$ and $\overline{X}$ represent the mean external nitrate and algal biomass levels for the time interval ΔT. The charge in external nitrate is represented by ΔN.

The scatter in the correlation plot may be indicative of a number of factors: (1) inappropriateness of Equation 2; (2) sensitivity of Equation 5 to low $\overline{N}$ and ΔN values; (3) sensitivity of utilizing linear interpolation for calculation of $\overline{N}$ and $\overline{X}$. Fortunately, the use of an essentially continuous simulation routine permits the elimination of factors 2 and 3, permitting the evaluation of Equation 2 for representing the dynamics of uptake.

A complete listing of the computer model used for calibration purposes is given in Appendix B. A fourth order Runge-Kutta algorithm was used to provide a numerical solution to Equations 1 through 3. A dilution routine was incorporated into the model to permit simulation of a sequence of dilutions. The external nitrate levels immediately after dilution were defined by the routine. The biomass was reduced automatically to 50 percent to reflect the effect of the 1:1 dilutions. These discontinuities were, of course, required by the external perturbations upon the system at the time of dilution. As a result of the nature of the dilution routine, errors generated in simulation of growth and uptake dynamics before dilution were transmitted in a continuous manner to phases after dilution.

Initial applications of the percent nitrogen model to the observed algal growth dynamics indicated the resolution of the percent nitrogen model was limited by the inability of Equation 1 to represent the growth

response to *S. capricornutum* immediately after the addition of fresh medium. The results of Experiment 1 in terms of external nitrate, percent nitrogen, and dry weight are shown in Figure 5. Immediately after dilution a lag in growth became apparent (4.73 - 5.05 days). Similar lags occurred after each dilution of Experiment 2 (Figure 6).

Previous authors have observed or described this phenomenon. The lag is explained when we examine the history of the culture in terms of nitrogen assimulation and storage patterns. Through the first phase of Experiment 1 (0.0 - 4.70 days) the nitrogen content decreases as the biomass increases (Daley and Brown, 1973). As the nitrogen content approaches the two percent level, the growth of the algae approaches zero. This decrease in percent nitrogen reflects the shift of nitrogen from intermediate and nonessential forms to forms essential for growth. At the 2 percent nitrogen level, the storage capability has been completely utilized. Growth at this point reflects continued carbohydrate and fat accumulation (Fogg, 1959; Vasconcelos and Fay, 1974).

Immediately after dilution (4.73 days) the cells begin the rapid uptake of the available nitrate. The nitrate is assimilated to organic intermediates, nonessential, and essential forms. As the cell synthesizes the necessary components (5.05 days), growth increases at a rapid pace (Vasconcelos and Fay, 1974).

According to this classical interpretation the lag in growth occurs as the cell "gears up" by synthesizing essential components. The response after dilution depends upon the past condition of the cell.

The basic assumption of Equation 1 is that a unique growth rate would be reflected for a given nutrient content. It is apparent from the results
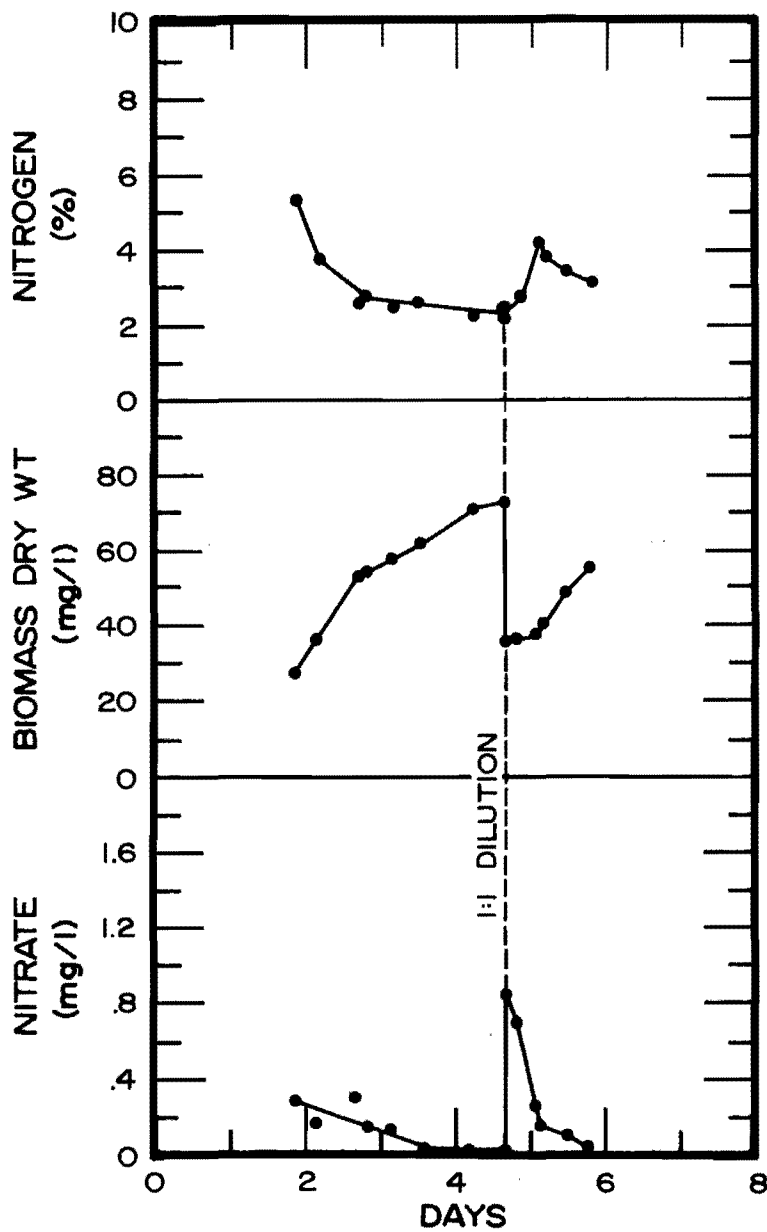
Figure 5.   Response of nitrogen deficient cultures to the addition of
nitrate during Experiment 1.
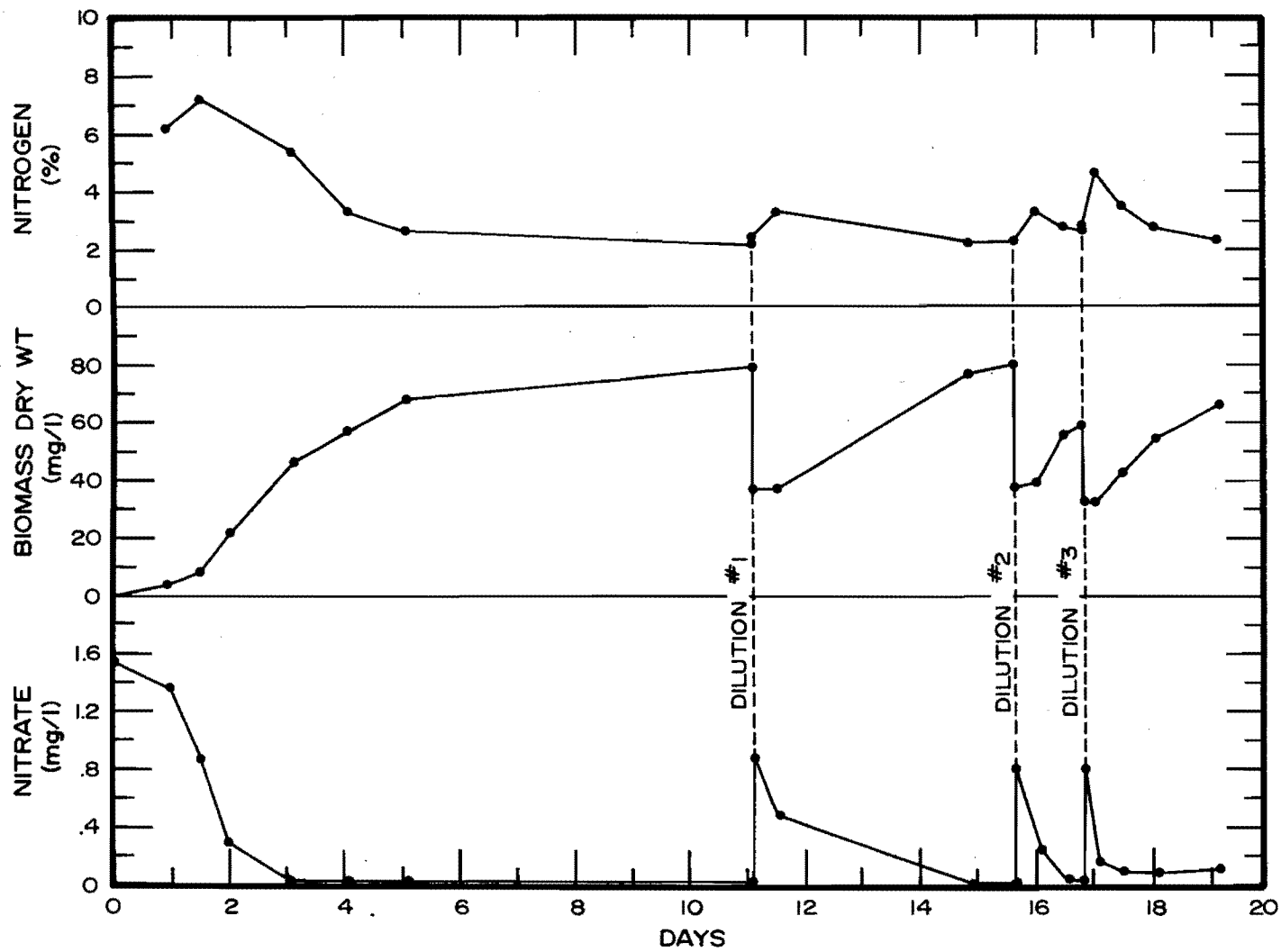
Figure 6. Responses of nitrogen deficient cultures to the addition of nitrate during Experiment 2.

that this assumption is not valid. Comparison of the growth responses for equivalent percent nitrogen values to the right and left of the percent nitrogen peaks in Figures 5 and 6 clearly demonstrate significant differences in growth responses. These preliminary observations would lead us to believe that internal nitrogen assimulation rates are not sufficiently fast to erase the effects of the cell history, leading to a unique percent nitrogen-growth rate correlation. Or put in other terms, the growth rate cannot be correlated to percent nitrogen without considering cell nutrient history. However, this conclusion is not necessarily correct.

Although Equation 1 does not apply to all situations its failure is not the result of slow response rates. In fact, the classical interpretation of lag does not explain the experimental data. The lag phenomenon is not solely the result of a depletion of internal nitrogen components as would be expected from the classical interpretation. This hypothesis is drawn largely from the response of the cultures after the third dilution of Experiment 2 (Figure 6). Despite substantial additional potential for growth, growth ceased upon the addition of nitrate. This strongly suggests that growth depends not only upon internal nitrogen levels but also upon external nitrogen availability. The availability of nitrate interrupts the normal pattern of nitrogen deficient growth causing a temporary discontinuation or reduction of growth.

The lag, therefore, could be explained in terms of a change in internal nitrogen pathways. Before the third dilution of Experiment 2, the cultures undoubtedly had entered the phase of utilization of higher nitrogen containing organic components to sustain growth. If the dilution

had not been performed, the cultures would have exhibited a slowly decreasing growth rate until the nitrogen stores were depleted. Upon the addition of nitrate to the system it would make little sense in terms of energy for a cell to continue to utilize the higher organic forms to provide nitrogen for components essential for growth. The experimental data indicate that the presence of nitrate in some manner inhibits growth possibly through the interruption of pathways of utilization of higher organics. The cell then continues the pathway of uptake and assimilation of nitrate while growth is interrupted.

The proposed mechanism provides a framework for modification of Equation 1. Examination of Figure 5 and Figure 6 reveals that in all cases, the end of the lag phase corresponds precisely with the percent nitrogen peaks. It appears that lag phase occurs while the percent nitrogen of the cell is increasing.

Figure 7 and Figure 8 represent correlation plots of specific growth rate versus percent nitrogen for phases of growth exhibiting decreasing and increasing percent nitrogen values respectively. It is immediately apparent that significantly different relationships exist for the two phases. Specific growth rates derived in periods with decreasing percent nitrogen correlate strongly with percent nitrogen ($r^2 = 0.883$) when a linear regression is utilized (Figure 7). This suggests that Equation 1 applies for periods of growth decreasing exhibiting percent nitrogen. The growth rate constant, U, and the minimum nitrogen level, NMIN, are defined by this correlation to be 0.39 and 2.11 respectively.

A plot of specific growth rate versus percent nitrogen for periods exhibiting increasing percent nitrogen is presented in Figure 8. A discontinuous response has been proposed for periods exhibiting increasing
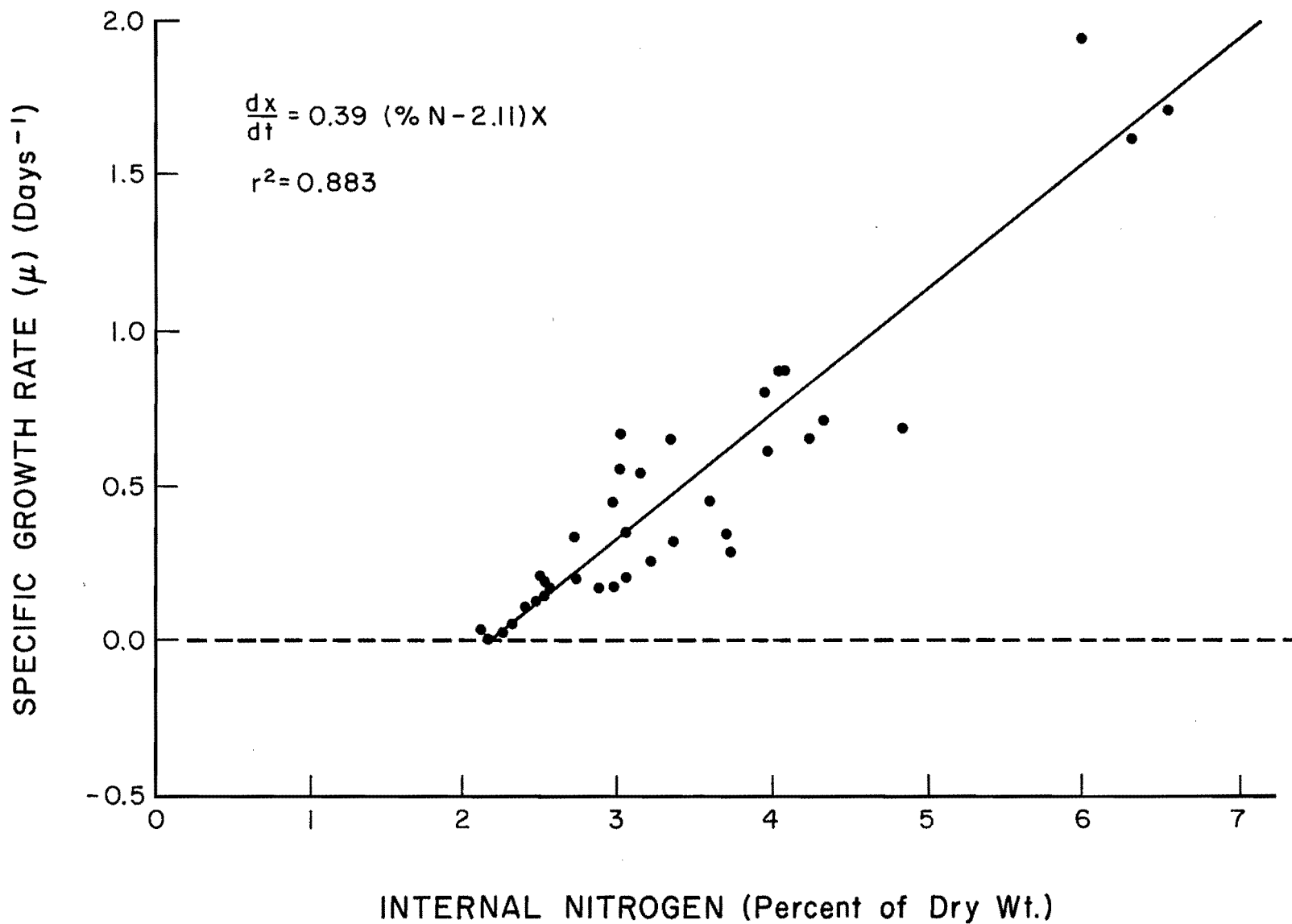
Figure 7.   Correlation of specific growth rate to mean percent nitrogen for phases of growth exhibiting decreasing nitrogen content.
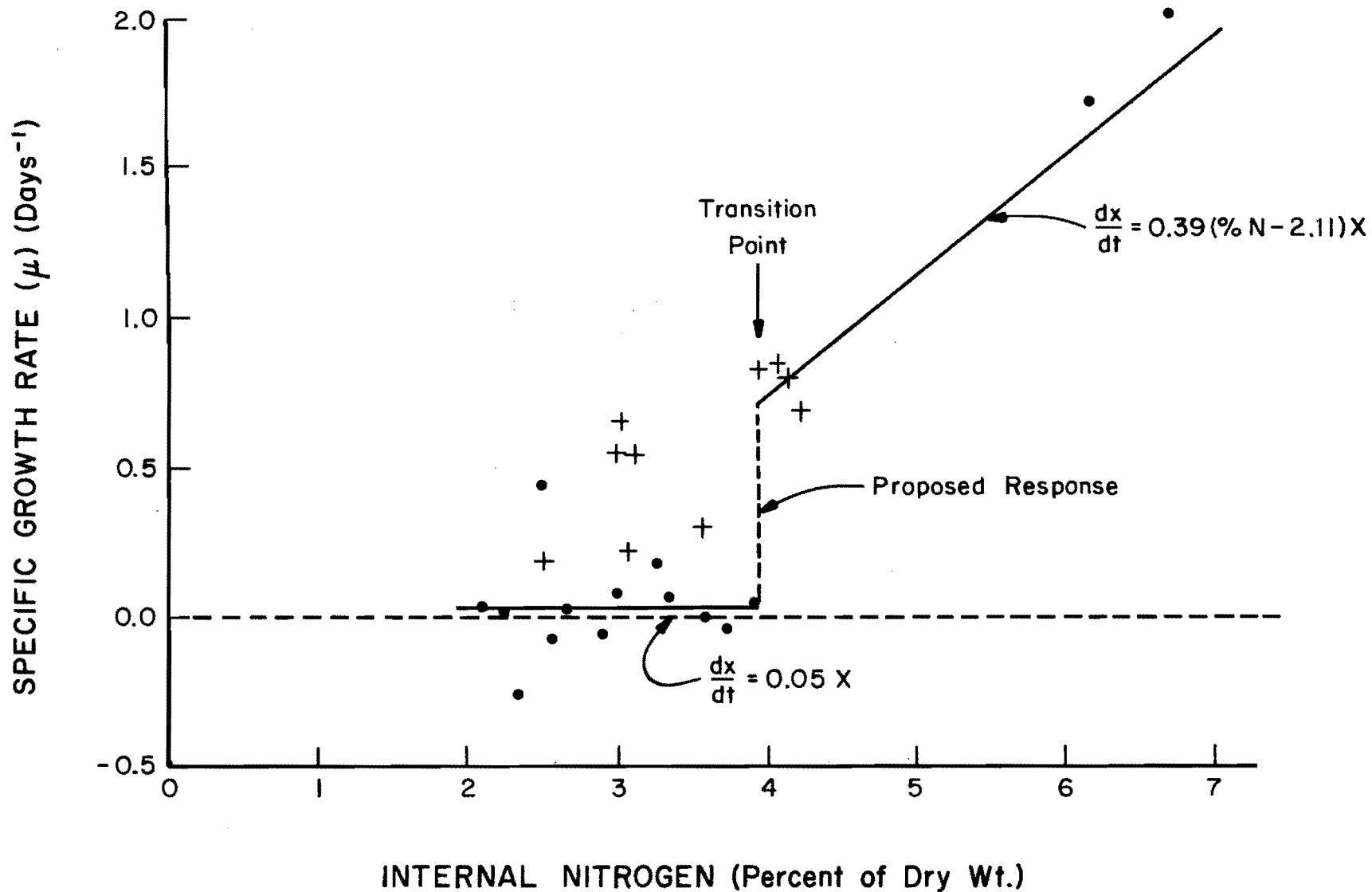
Figure 8. Correlation of specific growth rate to mean percent nitrogen for phases of growth exhibiting increasing nitrogen content.

percent nitrogen. For percent nitrogen values less than 3.8 percent, Equation 6 applies

$$\frac{dx}{dt} = U'x \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad (6)$$

in which

$U' = $ constant growth const.

Above 4 percent Equation 1 was assumed to apply. This function was se-lected since it is compatible with the observed correlation points (Figure 8) and the hypothesized growth inhibition mechanism. Above the 4 percent nitrogen level there are obviously insufficient observed data points to describe the functional response. This is undoubtedly in part an artifact of our experimental design. Uptake must more than compensate for growth if the percent nitrogen is to continue its increase. Under the experimental conditions existing immediately after dilution, the cultures were unable to obtain nitrogen content levels above 4.5 percent (Figure 6). As this level was approached rapid growth plus nitrate depletion in the medium led to a decreasing nitrogen content. If the nitrate availability was increased, the percent nitrogen may have con-tinued to increase during the period of high growth. Such a situation was observed early in Experiment 2 in the time interval from 0.93 days to 1.5 days (Figure 6). This time interval produced the two correlation points above 6 percent. The lack of correlation points between 4 and 6 percent reflects the failure of the experimental design to expose the cultures to the appropriate regimes of nitrate availability.

This gap can be filled in a reasonable manner if the proposed growth blocking mechanism is used as a framework. First, it was observed that the two correlation points above 6 percent on the increasing percent

nitrogen plot (Figure 8) fall close to the correlation line from the decreasing percent nitrogen plot (Figure 7). Secondly, it is only reasonable to assume that the rapid increase in growth causes the end of the increasing percent nitrogen regime. The magnitude of this growth rate is reflected by the initial (with respect to time) growth rates exhibited in the decreasing percent nitrogen regime. Since these points are also compatible with the correlation line from Figure 7, it is assumed that when the blocking mechanism is not in effect the growth response returns to normal. The normal distribution of internal nitrogen products has been re-established by the time growth begins.

The questions that remain are 1) how the transition from reduced growth to normal growth occurs and 2) where, with respect to percent nitrogen, does it occur. These questions cannot be answered without development of additional experimental data. Any conclusions drawn in this area would undoubtedly be seriously biased by the experimental design used here. It was, therefore, assumed that a discontinuous transition occurs at constant percent nitrogen level.

Figure 9 illustrates the relationship between the specific growth rate, μ, and percent nitrogen resulting from the application of Equations 3 and 6.

Plots comparing simulated curves with observed data resulting from application of the modified model to the results of Experiments 1 and 2 are illustrated in Figures 10 through 18. A single set of coefficients was utilized for these simulations. Experiment 2 was simulated in two segments (0.93 to 11.1 days, and 11.1 to 19.3 days) because of storage limits of the model. The Runge-Kutta Routine was applied with a 30 minute time step to all simulations.
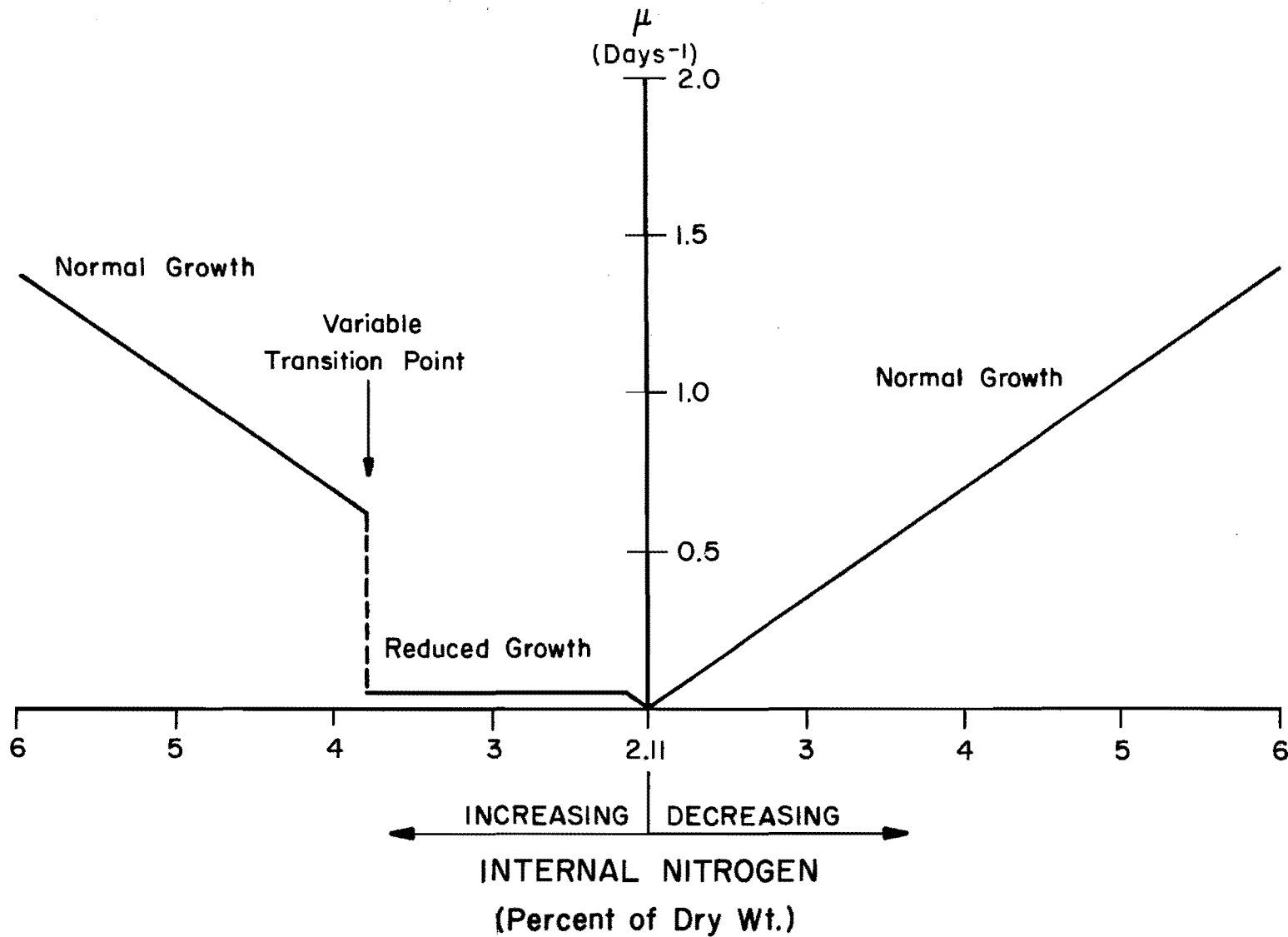
Figure 9.   Variation of specific growth rate with increasing and decreasing internal nitrogen contents.

SIMULATION OF ALGAL UPTAKE DYNAMICS BY PERCENT NITROGEN MODEL

```
          1.000  +--------------------+----------------+-----------------+-----------------+------------------+
                 !                                                                                          !
                 !                        0                                                                 !
                 !                                                                                          !
                 !                        0                                                                 !
                 !                        N                                                                 !
                 !                                                                                          !
                 !                        0                                                                 !
          0.7673 +                                                                                          +
                 !                        0                                                                 !
                 !                                                                                          !
   E             !                        ON                                                                !
   X             !                                                                                          !
   T             !                        0                                                                 !
   R             !                                                                                          !
   A             !                        0                                                                 !
   C             !                                                                                          !
   E  0.5347     +                         0                                                                +
   L             !                                                                                          !
   L             !                        0                                                                 !
   U             !                        0                                                                 !
   L             !                        0                                                                 !
   A             !                                                                                          !
   R             !                                                                                          !
                 !                          0                                                               !
   N             !                          0                                                               !
   I             !                          0                                                               !
   T  0.3020     +         N                0                                                               +
   R             !N                         0                                                               !
   O             !0                         0                                                               !
   G             !C                         NO                                                              !
   E             !0                          0                                                              !
   N             ! ON                        0                                                              !
                 ! 0       N  N              ON                                                             !
   M             ! 00                         0                                                             !
   G             ! 0                          0                                                             !
   /             ! 00                       OON                                                            !
   L  .6931E-01  +  0                         0                                                             +
                 ! 00                        DO                                                            !
                 !    00                     ODO                                                           !
                 !      0000000ONOCOONOOOO       ON                                                        !
                 !                                                                                          !
                 !                                                                                          !
                 !                                                                                          !
        -0.1634  +              N                                                                           +
                 +--------------------+----------------+-----------------+-----------------+------------------+
                1.833              4.233            6.633             9.033             11.43             13.83
                                              TIME (DAYS)
```

OBSERVED DATA INDICATED BY "N"
SIMULATED CURVE COMPOSED OF "C"
CONSTANTS FOR THIS RUN:AMIN=2.11;G=0.10;U=0.39
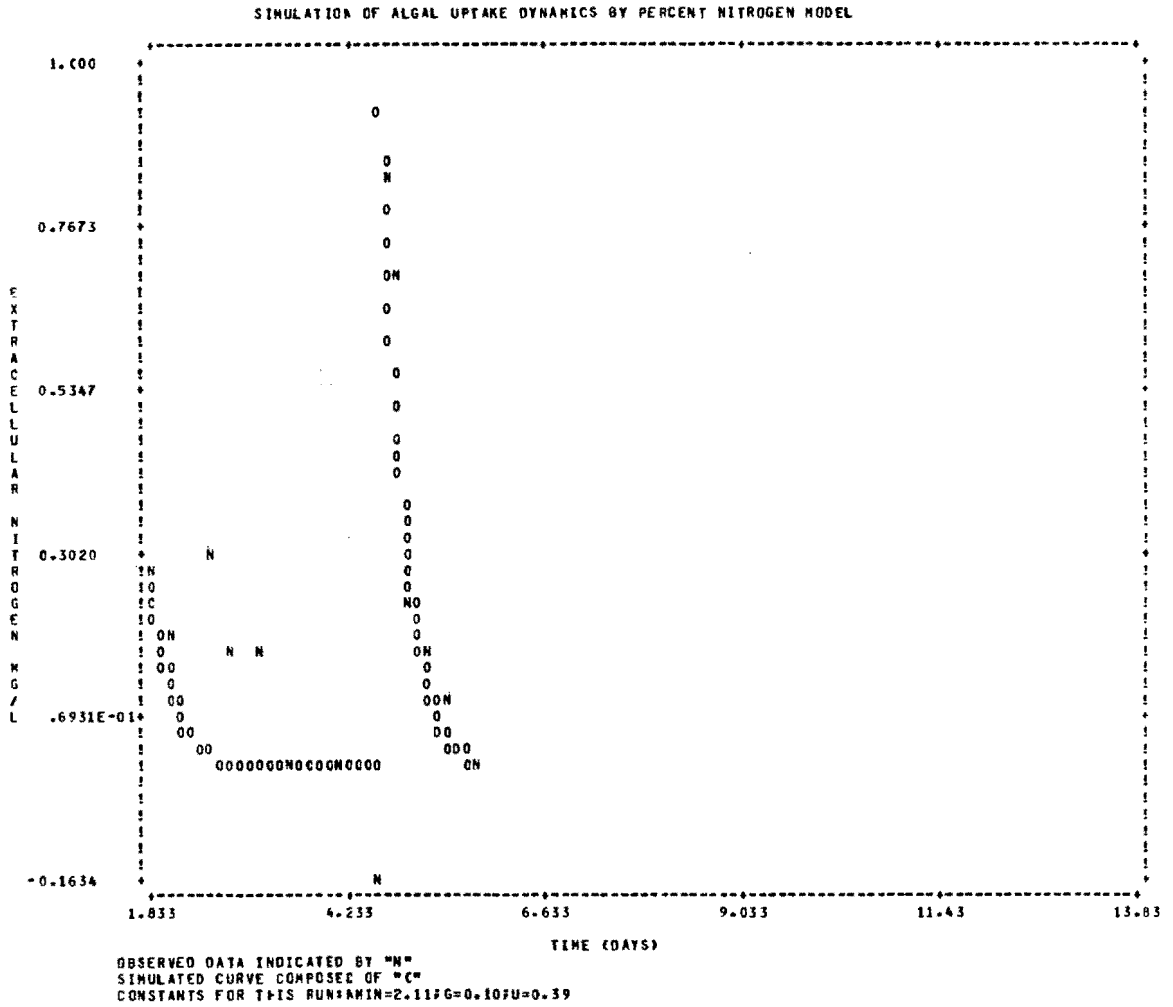
Figure 10. Comparison of simulated external nitrate curve with values
            calculated from Experiment 1, transition point = 3.8 percent.
            (See Appendix Table C-1.)

SIMULATION OF ALGAL GROWTH DYNAMICS BY PERCENT NITROGEN MODEL

```
100.0  +-------------------+-------------------+-------------------+-------------------+-------------------+-------------------+
       +                                                                                                                      +
       I                                                                                                                      I
       I                                                                                                                      I
       I                                                                                                                      I
       I                                                                                                                      I
       I                                                                                                                      I
 80.00 +                                                                                                                      +
       I                                                                                                                      I
       I                                                                                                                      I
       I                     X    X                                                                                           I
       I                    0 CO                                                                                              I
       I                  COO                                                                                                 I
       I                 COC                                                                                                  I
       I                OOC                                                                                                   I
       I               OX                                                                                                     I
 60.00 +           OO                                                                                                         +
D      I           OX                                                                                                         I
R      I          OO                                 O                                                                        I
Y      I         OO                                 OX                                                                        I
M      I       XOX                                  OO                                                                        I
T      I       O.                                   O                                                                         I
I      I      O                                                                                                              I
N      I     OO                                    OO                                                                         I
       I    OO                                      O                                                                         I
M      I    O                                      OX                                                                         I
G 40.00+   OO                                       O                                                                         +
/      I   O                                      XO                                                                          I
L      I  X                                    OXXO                                                                          I
       I OO                                                                                                                   I
       I O                                                                                                                    I
       IOO                                                                                                                    I
       IO                                                                                                                     I
       IX                                                                                                                     I
       I                                                                                                                      I
 20.00 +                                                                                                                      +
       I                                                                                                                      I
       I                                                                                                                      I
       I                                                                                                                      I
       I                                                                                                                      I
       I                                                                                                                      I
       I                                                                                                                      I
  0.   +-------------------+-------------------+-------------------+-------------------+-------------------+-------------------+
     1.833               4.233               6.633               9.033               11.43               13.83
                                              TIME (DAYS)
```

OBSERVED DATA INDICATED BY "X"
SIMULATED CURVE COMPOSED OF "C"
CONSTANTS FOR THIS RUN:AMIN=2.11;G=0.10;U=0.39
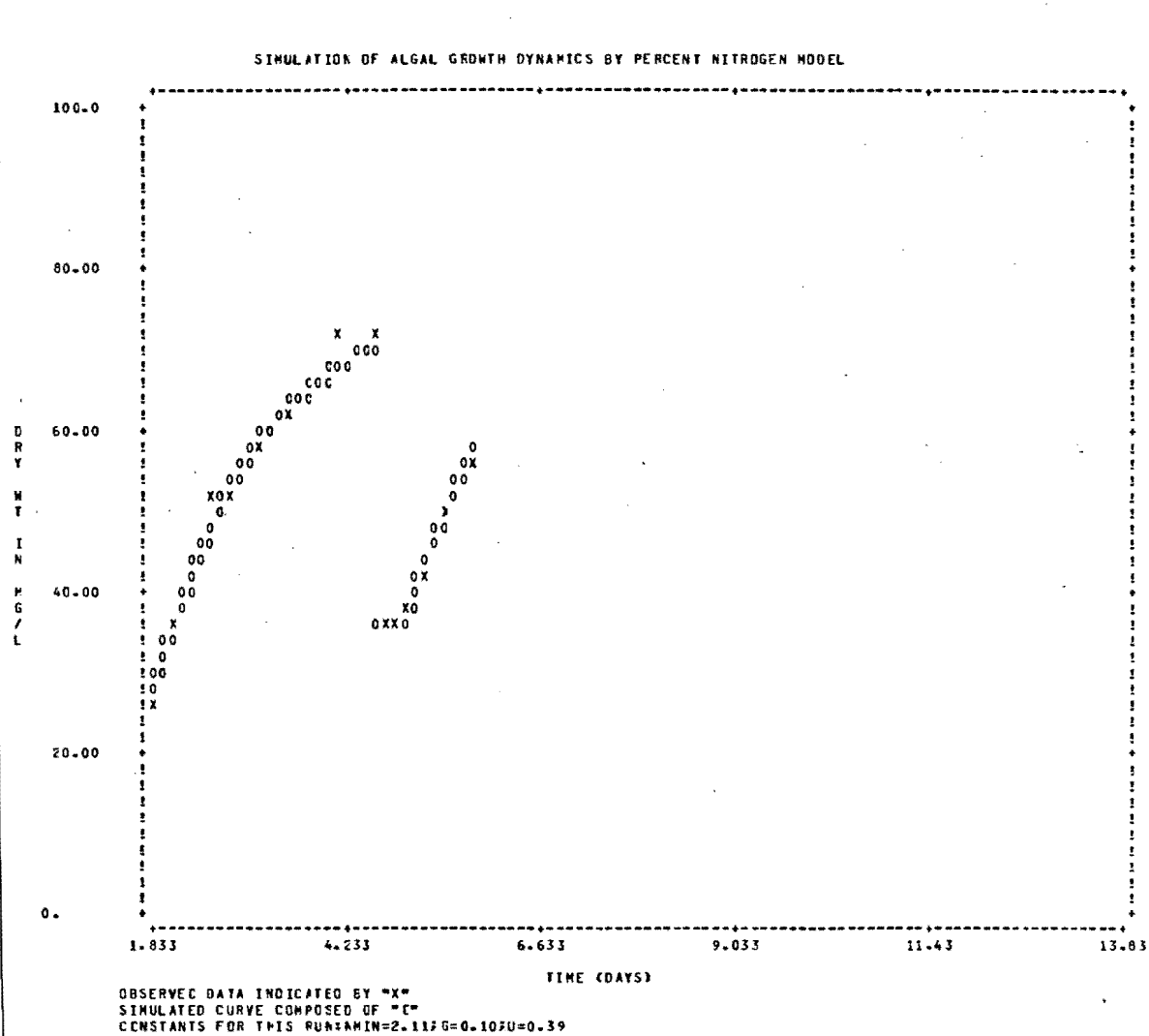
Figure 11. Comparison of simulated growth curve with points observed in Experiment 2, transition point = 3.8 percent. (See Appendix Table C-1.)
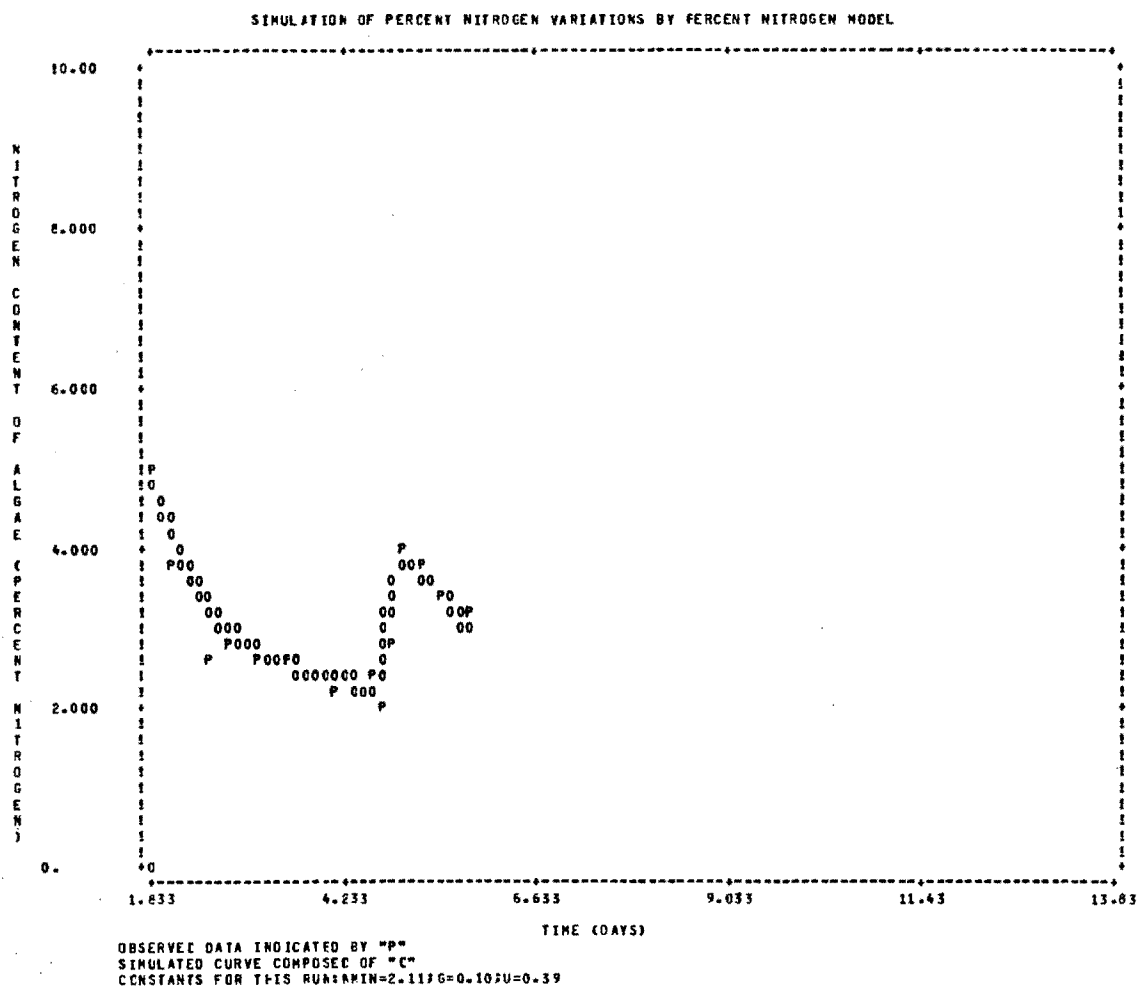
Figure 12. Comparison of simulated nitrogen content curve with observed percent nitrogen values from Experiment 1, transition point = 3.8 percent. (See Appendix Table C-1.)

SIMULATION OF ALGAL UPTAKE DYNAMICS BY PERCENT NITROGEN MODEL

```
         1.384  +N
                10
                100
                ! 0
                ! 00
                !  0
                !  0
                !   0
                !   0
                !   0
         1.084  +   00
                !   0
                !   0
                !    0
E               !    0
X               !    0
T               !    0
R               !  N 0
A               !    0
C               !    0
E    0.7846  +     0
L               !    0
L               !    0
U               !    0
L               !    0
A               !    C
R               !    C
                !    C
N               !    C
I               !    0
T    0.4848  +    0
R               !    0
O               !    0
G               !    0
E               !     0
N               !     0
                !  N  0
N               !    0
G               !    0
/               !    00
L    0.1851  +    0
                !    00
                !    0
                !    0
                !   00
                !   000
                !        N00 00 00 0N 00 00 00 00 0N 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00 00
                !
                !
        -0.1147  +                                                                                    N
                +----------------------+----------------------+----------------------+----------------------+----------------------+
              0.9300               3.330               5.730               8.130              10.53               12.93
                                              TIME (DAYS)
```

OBSERVED DATA INDICATED BY "N"
SIMULATED CURVE COMPOSED OF "O"
CONSTANTS FOR THIS RUN: AKIN=2.11, G=0.103, U=0.39

Figure 13. Comparison of simulated external nitrate curve with values
calculated from Experiment 2 before dilution, transition
point = 3.8 percent. (See Appendix Table C-2.)

SIMULATION OF ALGAL GROWTH DYNAMICS BY PERCENT NITROGEN MODEL

```
       100.0 +
                !
                !
                !
                !
                !
                !
        80.00 +                                                                    X
                !
                !                                              000000 00 00 0000 0000 00 00 00 00 0000 00 00 000
                !                                   000 000000
                !                              X0000
                !                            0000
                !                          0000
                !                        000
                !                       00
     D  60.00 +                     000
     R          !                   00  X
     Y          !                  00
                !                 00
     W          !                 0 0
     T          !                00
                !               X 0
     I          !               0
     N          !              00
                !             00
     M  40.00 +             0
     G          !            00
     /          !           0
     L          !          00
                !          0
                !         0
                !        00
                !       0
                !       0
                !     X  0
                !       00
        20.00 +        0
                !        00
                !       0 C
                !        0
                !       0
                !     X00
                !     000
                !   00
                ! X00
                !
         0. +
          +------------+------------+------------+------------+------------+
         0.9300       3.330        5.730        8.130        10.53        12.93
                                   TIME (DAYS)
```

OBSERVED DATA INDICATED BY "X"
SIMULATED CURVE COMPOSED OF "O"
CONSTANTS FOR THIS RUN: NMIN=2.11; G=0.10;U=0.39
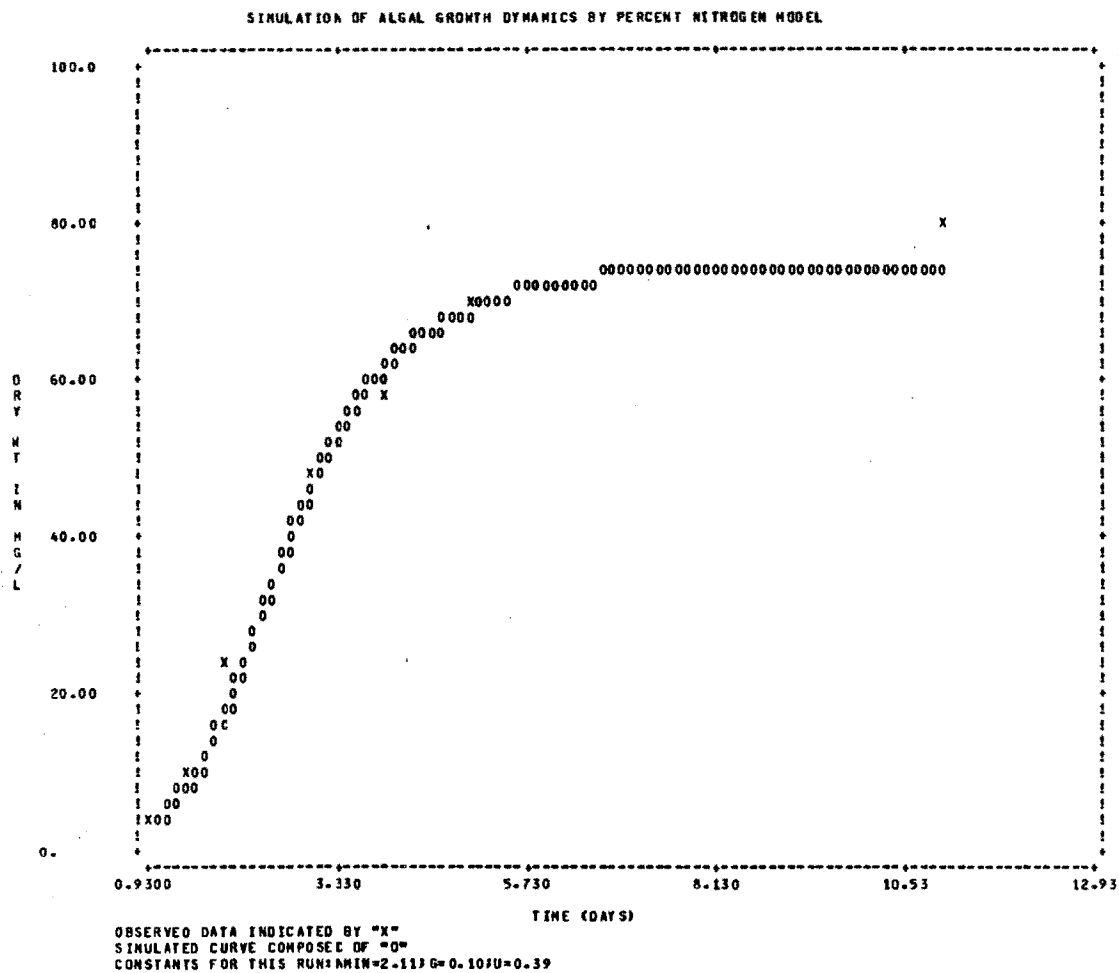
Figure 14. Comparison of simulated growth curve with points observed in Experiment 2 before dilution, transition point = 3.8 percent. (See Appendix Table C-2.)

SIMULATION OF PERCENT NITROGEN VARIATIONS BY PERCENT NITROGEN MODEL

OBSERVED DATA INDICATED BY "P"
SIMULATED CURVE COMPOSED OF "O"
CONSTANTS FOR THIS RUN: NNIN=2.11;G=0.107;U=0.39

Figure 15. Comparison of simulated nitrogen content curve with percent
nitrogen values observed in Experiment 2 before dilution,
transition point = 3.8 percent. (See Appendix Table C-2.)

SIMULATION OF ALGAL UPTAKE DYNAMICS BY PERCENT NITROGEN MODEL

```
          1.000  +----------------------+-----------------------+-----------------------+----------------------+----------------------+----------------------+
                 !                                                                                                                                            !
                 !                                                                                                                                            !
                 !                                                                                                                                            !
                 !                                                                                                                                            !
                 !                                                                                                                                            !
         0.7908  +                                                                                                                                            +
                 !N                                                                                                                                           !
                 !                                     N                                                                                                      !
    F            !O                                              N                                                                                            !
    X            !                                                                                                                                            !
    I            !O                              C        O                                                                                                   !
    R            !                                        O                                                                                                   !
    A            !C                              O        O                                                                                                   !
    C            !                                                                                                                                            !
    E    0.5817  +O                              C                                                                                                            +
    L            !                                        O                                                                                                   !
    L            !O                              C        O                                                                                                   !
    U            !                                        O                                                                                                   !
    L            ! O                             C                                                                                                            !
    A            !                                        O                                                                                                   !
    R            ! O  O                                   O                                                                                                   !
                 ! O N                           O        O                                                                                                   !
    N            ! O                             O        O                                                                                                   !
    I            ! O                             O        O                                                                                                   !
    T    0.3725  + O                             O         O                                                                                                  +
    R            ! O                                       O                                                                                                  !
    C            !   O                           O         O                                                                                                  !
    G            !   O                           O         O                                                                                                  !
    E            !   O                          O'         O                                                                                                  !
    N            !   O                          O         00                                                                                                  !
                 !   O                          O          O                                                                                                  !
    M            !   O                          O          O                                                                                                  !
    G            !   C                          O          O                                                                                                  !
    /            !   00                        ON          O                                                                                                  !
    L    0.1634  +   O                          O         0 C                                                                                                 +
                 !   O                          O          C                                                                                                  !
                 !    O                          O      N CO                                                                                                  !
                 !    O                          O          O                                                                                                 !
                 !     00                        00         00                    N                                                                           !
                 !    000       0 00 00CC CO CO C000 CO 0000 00 00 00 000 0  0000    N 0000   N0000000000                                                      !
                 !                                              N          ON         NO0 00 00 00 00                                                          !
      -.4579E-01+                                             N          N                                                                                    !
                 +----------------------+-----------------------+-----------------------+----------------------+----------------------+----------------------+
                 11.06                 13.46                   15.86                  18.26                  20.66                  23.06
```

TIME (DAYS)

OBSERVED DATA INDICATED BY "N"
SIMULATED CURVE COMPOSED OF "O"
CONSTANTS FOR THIS RUN:NMIN=2.11; G=0.10;U=0.35

Figure 16. Comparison of simulated external nitrate curve with values
calculated from the dilution sequence of Experiment 2, transition point = 3.8 percent. (See Appendix Table C-3.)

Figure 17. Comparison of simulated growth curve with points observed in
the dilution sequence of Experiment 2, transition point =
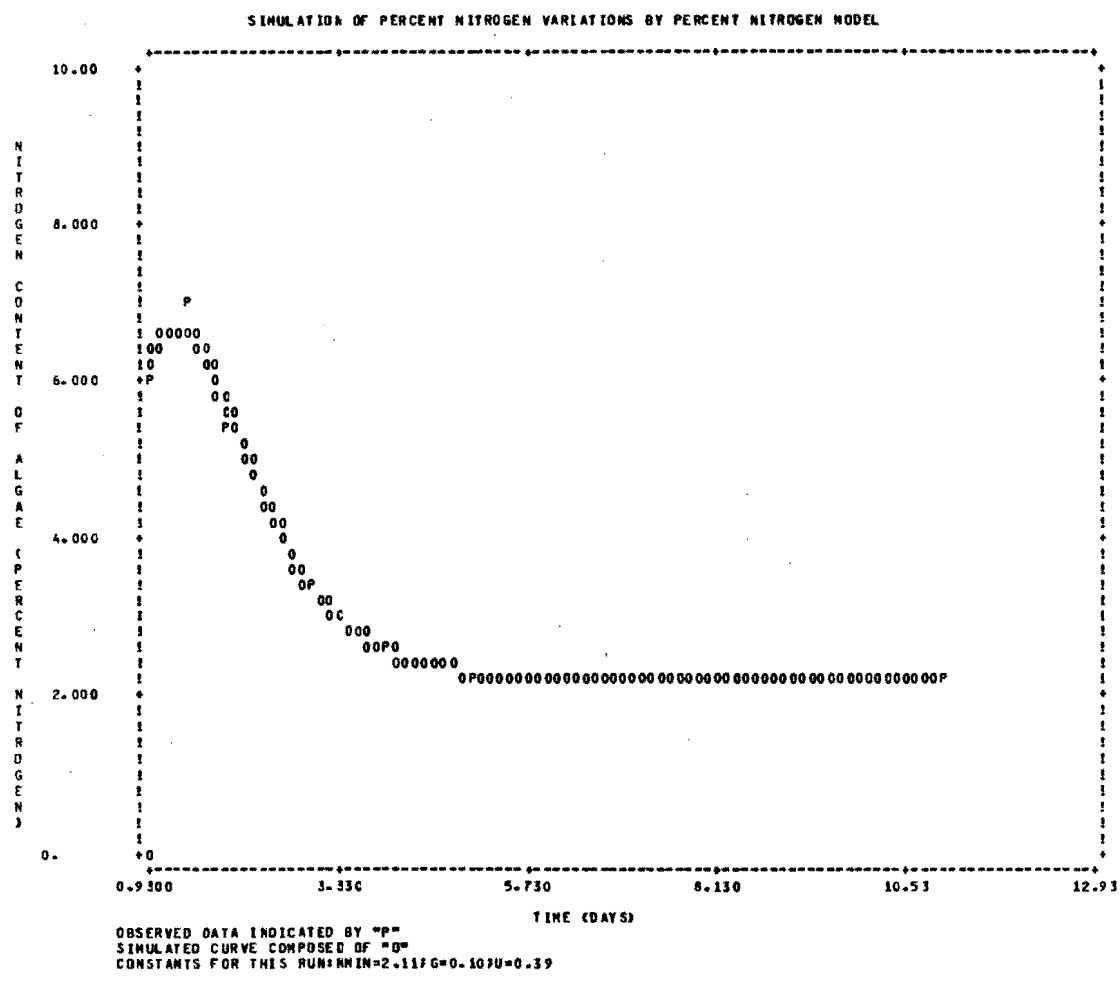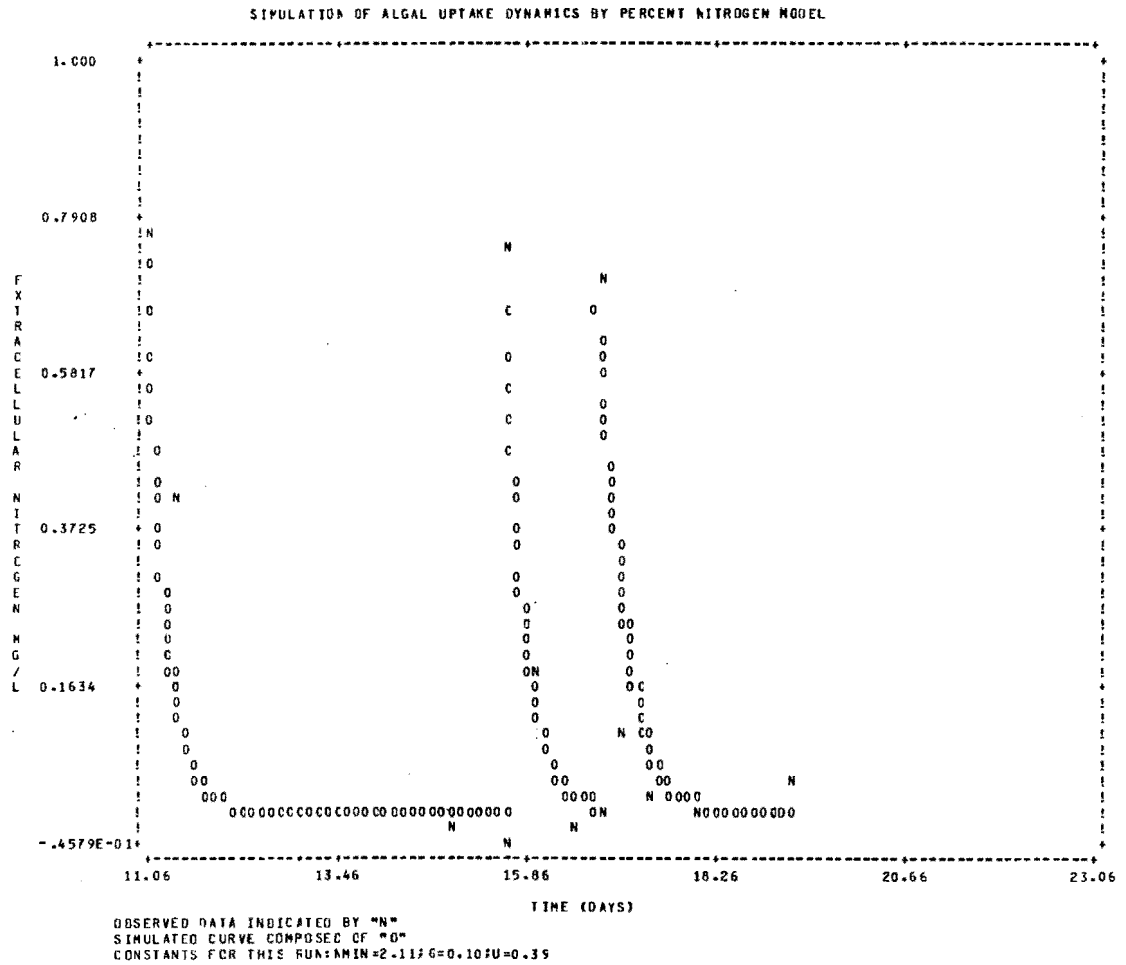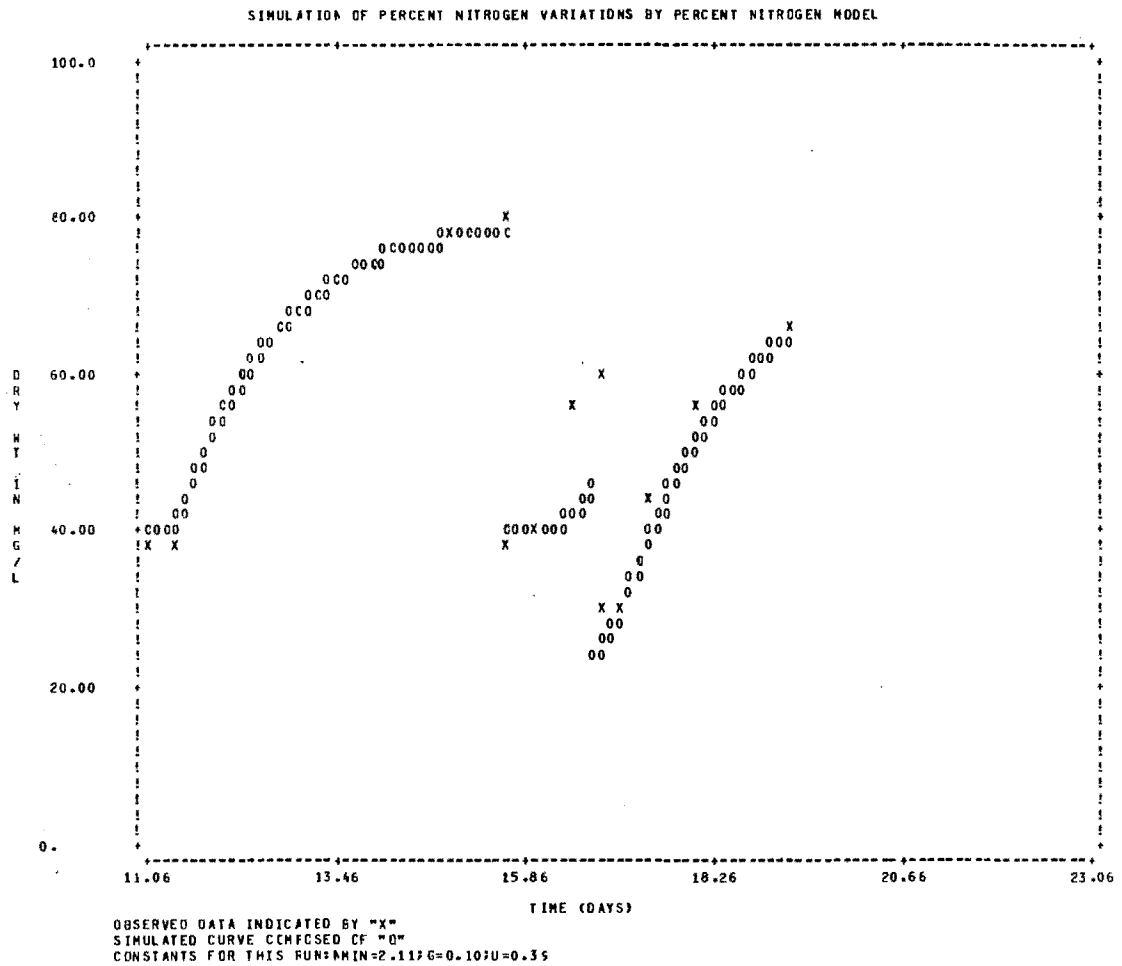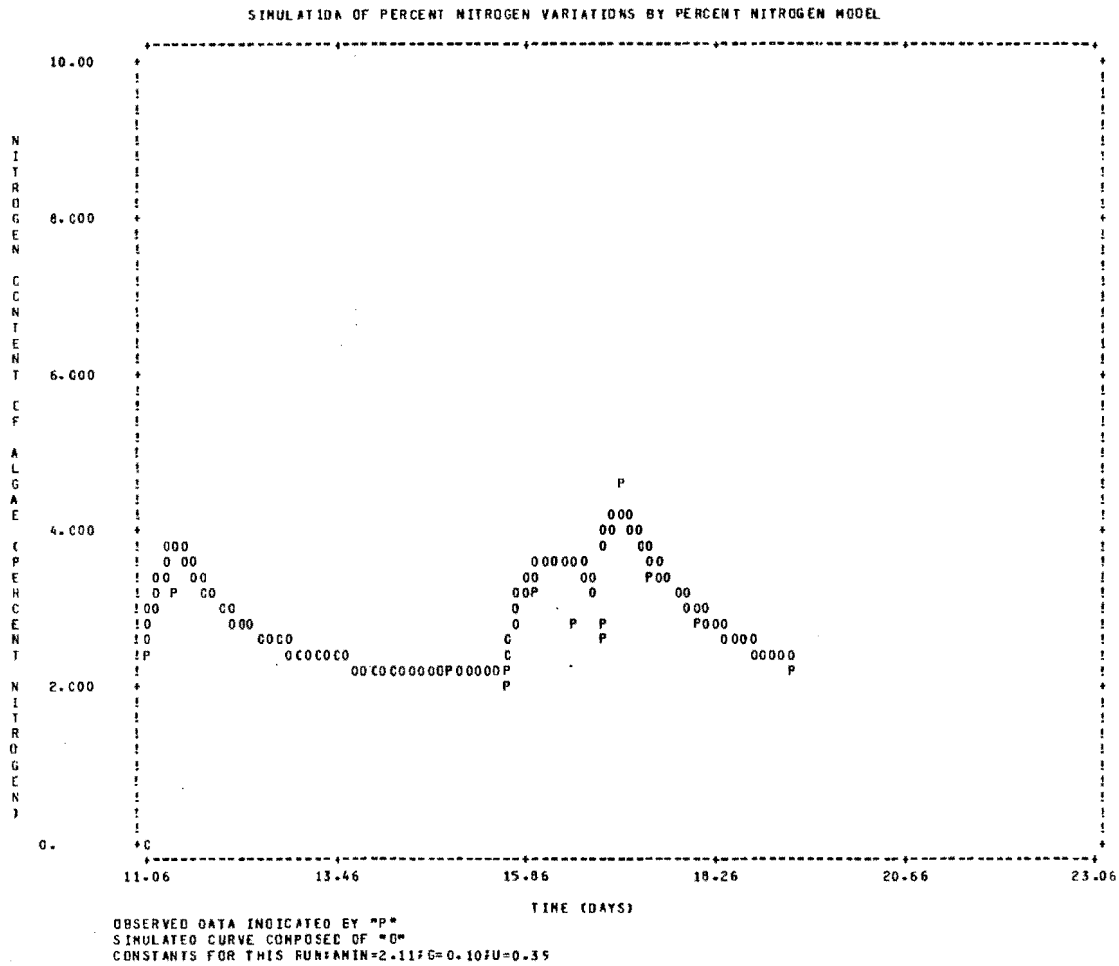3.8 percent. (See Appendix Table C-3.)

Figure 18. Comparison of simulated nitrogen content curve with percent
nitrogen values, observed in the dilution sequence of Experi-
ment 2, transition point = 3.8 percent.
(See Appendix Table C-3.)

The uptake rate constant, G, was selected by empirical methods to be 0.1 days^{-1}. This value provided good results when applied to the uptake data from Experiment 2 (Figures 13, 16, and 19). Negative data points result from the indirect calculation of the external nitrate levels. Application of Equation 2 to results of Experiment 1 revealed little correlation between predicted and observed points (Figure 10) for the period before dilution (4.70 days) and excellent correlation after dilution. The deviations before dilution are believed to have resulted from the variations in pH previously discussed. Equation 2 with an uptake constant of 0.1 days^{-1} provided satisfactory simulation of the uptake phenomenon observed.

Equation 1 was applied with constants derived from the regression analysis plot (Figure 7) to phases of growth exhibiting decreasing nitrogen contents. Equations 1 and 6 were applied to phases of increasing nitrogen contents. The transition point was found empirically to be 3.8 percent. The reduced growth rate constant, U', similarly determined to be 0.05 days^{-1}. The simulated percent nitrogen curves were calculated by the model from the simulated growth and uptake curves.

The most significant deviation of the model occurred between the second and third dilution of Experiment 2 (Figure 17, day 15.67 to 16.83). The model response reflects the selection of 3.8 percent as the transition point. This value provides excellent simulation of the lag phenomenon for the dilution of Experiment 1 (Figure 11) and the first dilution of Experiment 2. The simulated lag of the second dilution (Figure 11) exceeds the observed lag. A transition point of 3.0 percent provides a better representation of the lag phenomenon for the second diltuion, but fails to

represent the lag of the first dilution adequately (Figure 20). The optimum transition point for the third dilution occurs at a value above 4 percent. It is apparent that the transition point is not a constant.

Aside from this variability, the model provides excellent representation of the growth phenomenon. The minimum nitrogen content (2.11 percent) and the growth rate constant (0.39 days^{-1}) are correct for these experimental conditions. The simulated growth curves correspond closely with the observed data (Figures 11, 15, 17).

The percent nitrogen curves are presented in Figures 12, 15, and 21. Deviations in these curves are the result of accumulated errors in the growth and uptake simulations. The simulated curves closely represent trends in the storage response. Points of inflection are not represented precisely. In some cases this can be attributed to the variability of the transition point. The sensitivity to this parameter is revealed by comparison of Figure 18 to Fi ure 21 which represent transition points of 3.8 and 3.0 percent respectively. In other cases (Figure 15) deviations are attributed to small variations in the uptake curve (Figure 13) during periods of rapid uptake. Fortunately, the growth model is totally insensitive to these minor deviations.

SIMULATION OF ALGAL UPTAKE DYNAMICS BY PERCENT NITROGEN MODEL
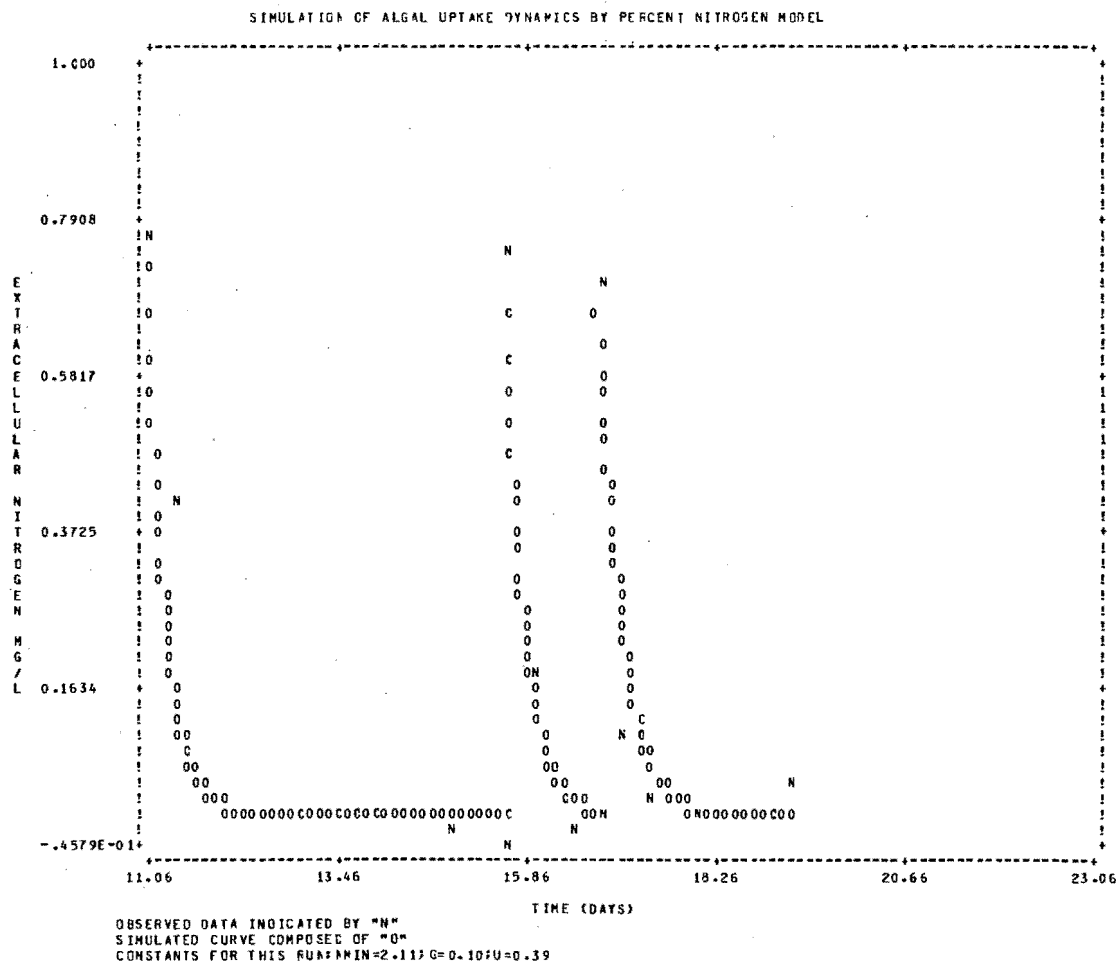
Figure 19. Comparison of simulated external nitrate curve with values calculated from the dilution sequence of Experiment 2, transition point = 3.8 percent. (See Appendix Table C-4.)

Figure 20. Comparison of simulated growth curve with points observed in the dilution sequence of Experiment 2 transition point modified to 3.0 percent. (See Appendix Table C-4.)
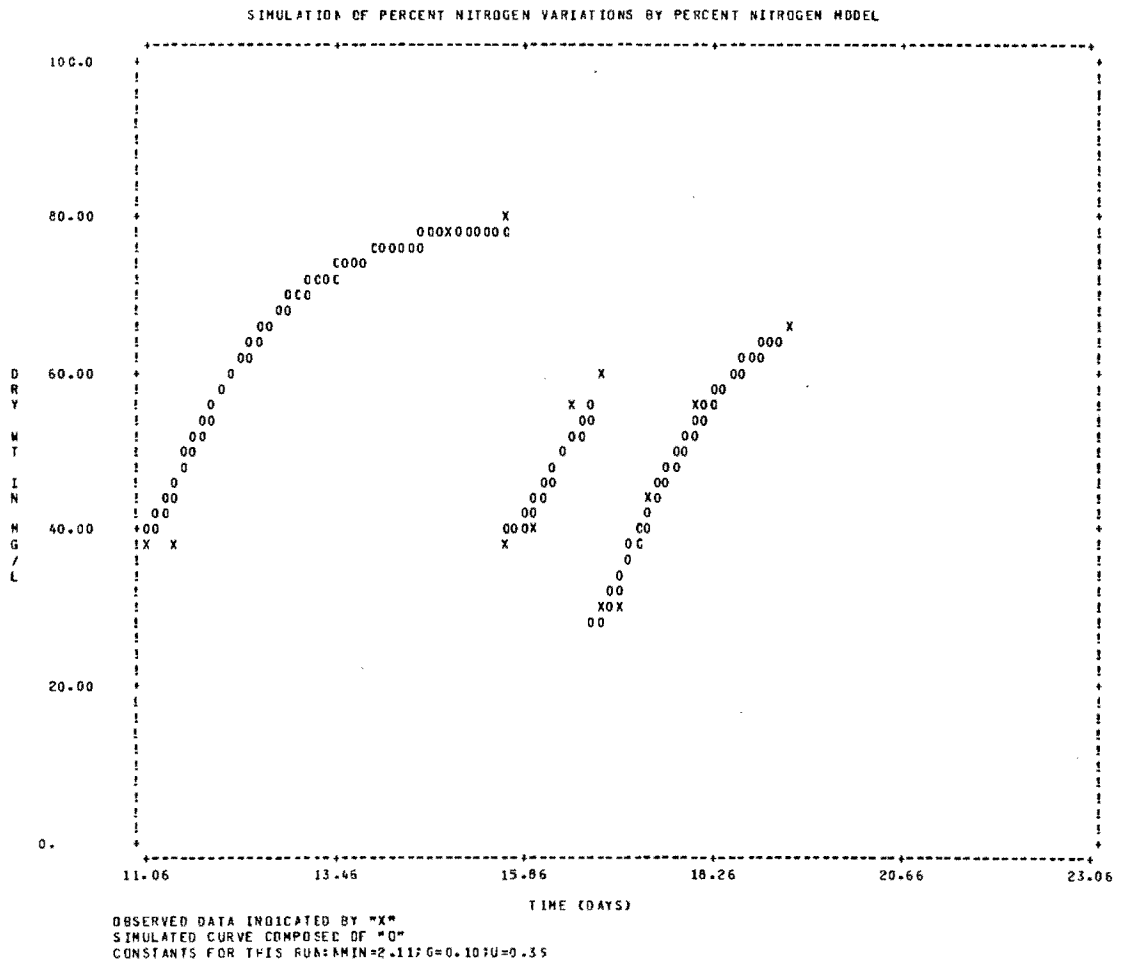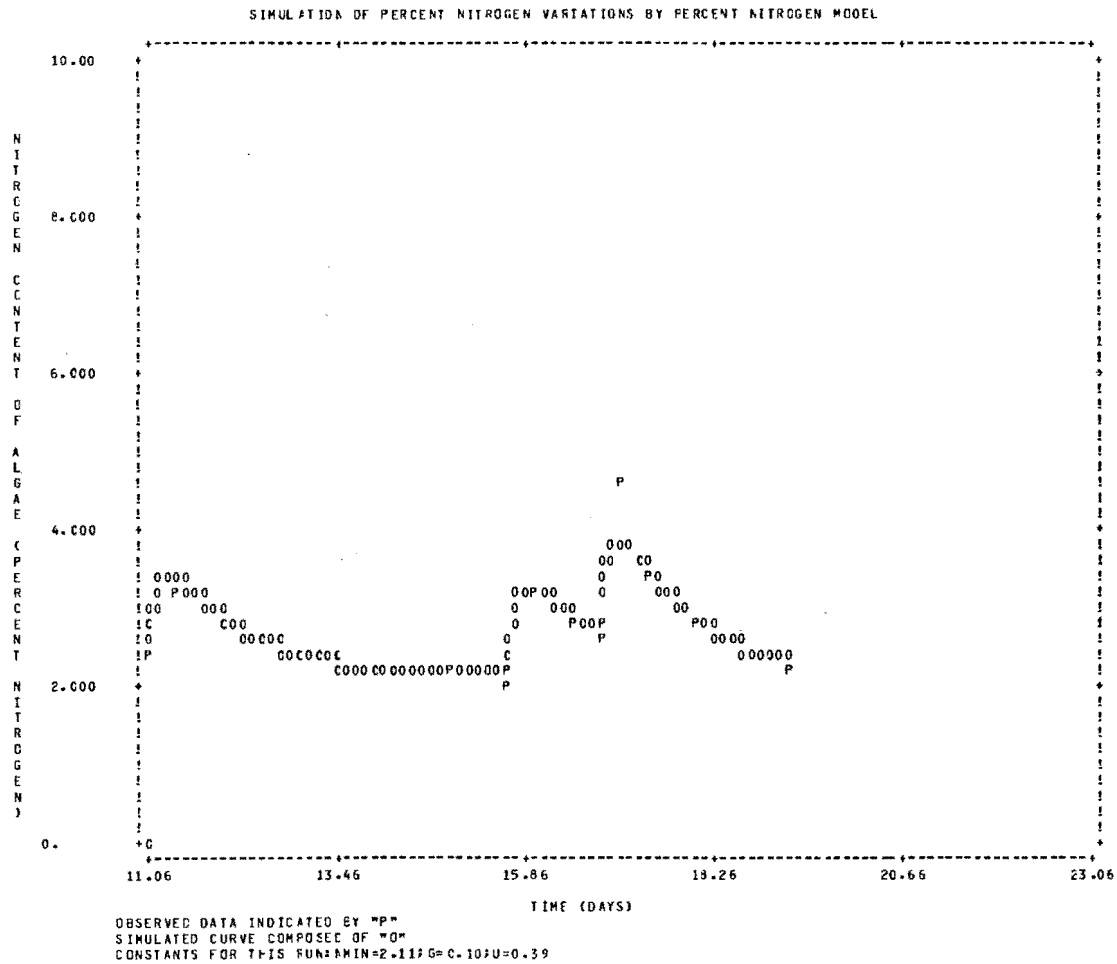
Figure 21. Comparison of simulated nitrogen content curve with percent nitrogen values observed in the dilution sequence of Experiment 2, transition point modified to 3.0 percent. (See Appendix Table C-4.)

SUMMARY

The mathematical model developed to represent algal uptake and growth phenomenon was based upon two underlying assumptions. First it was assumed that under a given set of environmental conditions cell components were synthesized or depleted in an established order. Secondly, it was assumed that the internal reaction rates were fast. Fast reaction rates would assure that a given nutrient content would reflect a given physiological state.

A conceptual model distinguishing between essential and nonessential nitrogen forms was utilized to represent algal storage phenomenon. Correlation studies indicated that uptake phenomenon was independent of internal nitrogen levels within the regime of nitrogen limitation studied. Equation 1 was found to provide satisfactory results when applied with an uptake constant of $G = 0.1$ days^{-1}. Minor deviations from the observed data (Figures 10, 13, and 16) plus poor correlation (Figure 4) suggest refinement in this area may be warranted. Deviations resulting from the application of Equation 1, however, did not affect growth simulations.

A linear relationship ($r^2 = 0.817$) was indicated between the specific growth rate and internal nitrogen levels of batch cultures of S. *capricornutum*. Application of this relationship in the form of Equation 2 revealed consistent deviation of the model during lag phases of growth. Observation revealed that lag phases always occurred during phases displaying low and increasing levels of internal nitrogen. By identifying those phases of growth exhibiting a decreasing internal nitrogen percentage and correlating only related points the linear relationship was

improved ($r^2 = 0.883$). From this regression analysis the constants for

Equation 2 were defined. The minimum percent nitrogen was found to be

2.11 percent and the growth rate constant, U, 0.39 days^{-1}. These con-

stant were used to describe "normal" growth since the high correlation

suggested that the underlying assumptions applied completing during peri-

ods of decreasing nitrogen content.

Analysis of the data related to increasing internal nitrogen levels

suggested a discontinuous response reflecting a reduced growth phase (lag)

and a normal growth phase. The reduced growth phase was explained by an

apparent violation of the second underlying assumption of the model.

During periods of rapid nutrient uptake and low internal nitrogen levels

growth rates did not correlate with internal nitrogen contents. Instead

a low (U = 0.05 days^{-1}) and constant growth rate was displayed. The

response of spiked cultures suggested the presence of a growth inhibition

mechanism during periods of rapid uptake. The algal model was modified

to exhibit a reduced growth phase with a constant specific growth rate

and a normal growth phase. The transition between reduced and normal

growth occurred at a variable nitrogen content in the range of 3 to 4.5

percent nitrogen on the increasing curve.

Application of the modified growth model displayed exellent results.

The model was insensitive to error accumulation. Growth rates and ulti-

mate biomasses were closely approximated. The model was found to be

sensitive to the selection of the transition point which was assumed con-

stant for the simulations. Simulated curves, however, corresponded

closely with observed data when the appropriate transition point was

selected. This indicated that the equations were valid. Factors

affecting the transition point could not be investigated without develop-

ment of additional data.

CONCLUSIONS AND RECOMMENDATIONS

1) There is a strong correlation between internal nitrogen levels and specific growth rate in batch cultures of *Selenastrum capricornutum*.

2) The uptake phenomenon was independent of the nitrogen content of algal cells for the regime of nitrogen limitation studied.

3) The data indicate the existence of a growth inhibition mechanism during periods of low increasing percent nitrogen. Factors contributing to the transition between reduced and normal growth are not clear.

It is believed that a number of areas related to nutrient content models warrants further study. The following are recommended:

1) Investigation of the factors affecting the proposed growth inhibition mechanism.

2) Expansion of the model to include phosphorus limited growth.

3) Sensitivity studies to examine variability of constants with environmental factors such as light and temperature.

4) Application of the model to other algal species to determine universality of relationships.

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APPENDICES

Appendix A

Data Listings for Experiments 1 and 2

Table A-1. Data listings for experiment 1. (Zero indicates no observation.)

EXPERIMENT #1,FLASK R,10/31/75,N=1.44

TIME (DAYS)	PH	TEMP DEG C	DO	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
0.60	7.02	26.20	.000	0.	0.00	0.00	0.000	TEMP AND CO2 ADJUSTED
1.71	9.24	26.70	.000	0.	0.00	0.00	0.000	SAME,BUBBLING AND MIXING INCREASED
1.83	7.51	26.40	.069	.132E+07	45.80	28.10	4.776	TEMP AND CO2 ADJUSTED
2.10	7.75	24.50	.102	0.	0.00	36.87	4.004	AIR OFF,TEMP AND CO2 ADJUSTED
2.67	6.49	24.00	.144	.356E+07	37.40	50.60	2.366	CO2 ADJUSTED
2.85	6.82	24.40	.151	.369E+07	37.20	51.80	2.594	CO2 ADJUSTED
3.16	6.70	23.60	.167	0.	0.00	58.40	2.586	CO2 ADJUSTED
3.55	7.42	24.20	.187	.405E+01	36.80	62.20	2.466	CO2 ADJUSTED
4.18	7.30	23.90	.197	0.	0.00	68.60	2.358	
4.70	7.32	24.00	.212	.433E+01	38.30	73.20	2.673	SETTLING,MIXING BAR OFF

EXPERIMENT #1,FLASK C,10/31/75,N=1.44

TIME (DAYS)	PH	TEMP DEG C	DO	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
0.60	7.10	27.20	.000	0.	0.00	0.00	0.000	TEMP AND CO2 ADJUSTED
1.71	9.95	27.50	.000	0.	0.00	0.00	0.000	SAME,NO AIR,BUBBLING-MIXING INCREASED
1.83	7.71	27.20	.057	.950E+06	46.70	22.80	5.538	TEMP AND CO2 ADJUSTED
2.10	7.80	25.60	.096	0.	0.00	33.85	4.037	TEMP AND CO2 ADJUSTED
2.67	6.49	25.00	.142	.336E+07	34.80	0.00	0.000	CO2 ADJUSTED
2.85	6.85	25.30	.154	.356E+07	33.40	50.40	3.062	CO2 ADJUSTED
3.16	6.70	24.90	.169	0.	0.00	55.60	2.461	CO2 ADJUSTED
3.55	7.36	25.50	.184	.422E+01	35.10	60.60	2.656	CO2 ADJUSTED
4.18	7.28	25.10	.196	0.	0.00	73.00	2.177	
4.70	7.30	25.00	.208	.442E+01	36.50	71.40	2.199	SETTLING,MIXING BAR OFF

EXPERIMENT #1,FLASK L,10/31/75,N=1.44

TIME (DAYS)	PH	TEMP DEG C	DO	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
0.60	7.00	26.70	.000	0.	0.00	0.00	0.000	TEMP AND CO2 ADJUSTED
1.71	9.16	26.80	.066	0.	0.00	0.00	0.000	SAME,NO AIR,BUBBLING-MIXING INCREASED
1.83	7.67	26.00	.071	.145E+07	42.50	27.80	4.937	TEMP AND CO2 ADJUSTED
2.10	7.70	24.50	.108	0.	0.00	38.90	3.566	TEMP AND CO2 ADJUSTED
2.67	6.49	23.60	.141	.326E+07	37.00	51.80	2.691	CO2 ADJUSTED
2.85	6.83	24.20	.154	.334E+07	37.40	52.60	2.736	CO2 ADJUSTED
3.16	6.70	23.60	.171	0.	0.00	59.00	2.486	CO2 ADJUSTED
3.55	7.40	24.60	.184	.407E+01	37.80	61.40	2.686	CO2 ADJUSTED
4.18	7.29	24.20	.200	0.	0.00	73.20	2.186	
4.70	7.30	24.00	.210	.425E+01	37.00	0.00	0.000	SETTLING,MIXING BAR OFF

Table A-1. Continued.

EXPERIMENT #1,FLASK R,AFTER DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
4.73	0.00	0.00	.106	.212E+07	37.20	37.40	2.175	CULTURE MIXED 50-50 WITH FRESH MEDIA
4.88	7.30	23.40	.105	.215E+07	37.60	36.00	2.480	
.05	7.30	24.00	.106	.215E+07	37.60	36.40	4.197	
.20	0.00	0.00	.000	.217E+07	44.30	41.40	3.931	
.52	0.00	0.00	.000	.262E+07	44.40	49.60	0.000	SETTLING,MIXING BAR OFF
81	7.20	24.00	.169	0.	0.00	54.40	3.236	SETTLING,MIXING BAR OFF

EXPERIMENT #1,FLASK C,AFTER DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
4.73	0.00	0.00	.109	.222E+07	35.50	36.40	2.001	CULTURE MIXED 50-50 WITH FRESH MEDIA
4.88	0.00	24.50	.107	.220E+07	35.50	36.00	3.086	
5.05	7.40	0.00	.107	.222E+07	36.40	35.80	4.320	
5.20	0.00	0.00	.000	.224E+07	41.70	40.80	3.789	
5.52	0.00	0.00	.000	.261E+07	45.20	45.80	3.582	SETTLING,MIXING BAR OFF
5.81	7.40	25.80	.170	0.	0.00	56.00	3.041	

EXPERIMENT #1,FLASK L,AFTER DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
4.73	0.00	0.00	.104	.209E+07	37.50	35.40	2.084	CULTURE MIXED 50-50 WITH FRESH MEDIA
.88	7.24	23.50	.107	.211E+07	37.70	37.80	2.814	
.05	0.00	24.00	.109	.211E+07	38.10	39.00	3.636	
.20	0.00	0.00	.000	.207E+07	42.60	40.80	3.734	
.52	0.00	0.00	.000	.234E+07	46.50	51.60	0.000	SETTLING,MIXING BAR OFF
.81	7.32	24.60	.168	0.	0.00	0.00	0.000	

Table A-2. Data listings for experiment 2. (Zero indicates no observation.)

EXPERIMENT #2,FLASK R,AT START

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
0.00	6.95	24.00	.000	0.	0.00	0.00	5.662	
0.93	7.05	24.00	.008	0.	0.00	3.40	5.376	
1.50	6.85	24.00	.021	0.	0.00	9.00	7.008	CO2 ADJUSTED
2.00	7.00	24.00	.059	0.	0.00	21.30	6.014	
3.05	6.90	24.40	.140	0.	0.00	44.40	3.632	BICARBONATE SOL IN AIR STRIPPING SERIES
4.03	7.00	24.00	.180	0.	0.00	57.60	2.790	
5.01	6.90	24.00	.200	0.	0.00	70.60	2.195	
11.04	7.00	24.50	.244	0.	0.00	79.60	2.137	

EXPERIMENT #2,FLASK C,AT START

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
0.00	7.00	24.00	.000	0.	0.00	0.00	5.662	
0.93	7.15	25.00	.011	0.	0.00	3.06	6.179	
1.50	6.95	25.00	.023	0.	0.00	9.90	7.243	CO2 EXHAUSTED
2.00	7.20	26.00	.067	0.	0.00	25.70	4.695	
3.05	7.20	26.00	.152	0.	0.00	49.00	3.256	BICARBONATE SOL IN AIR STRIPPING SERIES
4.03	7.05	25.00	.190	0.	0.00	59.20	2.650	
5.01	6.90	26.00	.210	0.	0.00	68.00	2.196	
11.04	7.10	26.00	.235	0.	0.00	77.20	2.225	

EXPERIMENT #2,FLASK L,AT START

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
0.00	7.10	24.00	.000	0.	0.00	0.00	5.662	
0.93	7.15	25.00	.012	0.	0.00	2.94	0.000	
1.50	7.10	25.00	.024	0.	0.00	10.40	6.983	CO2 ADJUSTED
2.00	7.50	25.00	.064	0.	0.00	23.20	5.532	
3.05	7.20	25.00	.147	0.	0.00	48.60	3.067	BICARBONATE SOL IN AIR STRIPPING SERIES
4.03	7.10	25.00	.192	0.	0.00	57.80	2.616	
5.01	7.00	25.00	.217	0.	0.00	69.20	2.416	
11.04	7.00	25.50	.239	0.	0.00	80.20	2.039	

Table A-2. Continued.

EXPERIMENT #2,FLASK R,AFTER 1ST DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
11.06	7.30	22.00	.126	0.	0.00	39.40	2.497	AFTER 50-50 DILUTION WITH FRESH MEDIUM
11.52	7.10	24.50	.131	0.	0.00	0.00	0.000	
14.97	7.10	24.00	.242	0.	0.00	78.40	1.991	
15.64	7.10	24.00	.254	0.	0.00	80.20	2.200	

EXPERIMENT #2,FLASK C,AFTER 1ST DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
11.06	7.35	22.50	.118	0.	0.00	38.40	2.324	AFTER 50-50 DILUTION WITH FRESH MEDIUM
11.52	7.10	25.50	.120	0.	0.00	39.00	2.914	
14.97	7.10	25.60	.236	0.	0.00	77.20	2.189	
15.64	7.20	25.50	.242	0.	0.00	79.40	2.090	

EXPERIMENT #2,FLASK L,AFTER 1ST DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
11.06	7.20	22.70	.124	0.	0.00	39.00	2.115	AFTER 50-50 DILUTION WITH FRESH MEDIUM
11.52	7.05	25.50	.130	0.	0.00	38.20	3.634	
14.97	7.10	25.20	.242	0.	0.00	77.40	2.415	
15.64	7.05	25.20	.250	0.	0.00	80.40	2.181	

Table A-2. Continued.

EXPERIMENT #2,FLASK R,AFTER 2ND DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
15.67	7.30	24.20	.132	0.	0.00	38.60	1.962	AFTER 50-50 DILUTION WITH FRESH MEDIUM
16.05	7.15	24.00	.132	0.	0.00	41.40	3.340	
16.56	7.13	24.50	.163	0.	0.00	55.20	2.850	
16.83	7.10	24.60	.180	0.	0.00	58.60	2.664	

EXPERIMENT #2,FLASK C,AFTER 2ND DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
15.67	7.25	25.30	.126	0.	0.00	38.00	2.030	AFTER 50-50 DILUTION WITH FRESH MEDIUM
16.05	7.10	25.50	.129	0.	0.00	40.20	3.290	
16.56	7.12	25.80	.162	0.	0.00	56.60	2.730	
16.83	7.11	26.00	.182	0.	0.00	59.60	2.426	

EXPERIMENT #2,FLASK L,AFTER 2ND DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
15.67	7.20	25.20	.125	0.	0.00	39.60	2.090	AFTER 50-50 DILUTION WITH FRESH MEDIUM
16.05	7.10	25.70	.130	0.	0.00	41.20	3.250	
16.56	7.05	25.20	.164	0.	0.00	54.60	2.810	
16.83	7.05	26.00	.184	0.	0.00	60.00	2.639	

Table A-2. Continued.

EXPERIMENT #2,FLASK R,AFTER 3RD DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
16.85	7.30	24.10	.094	0.	0.00	30.00	2.849	AFTER 50-50 DILUTION WITH FRESH MEDIUM
17.06	7.14	24.50	.095	0.	0.00	30.30	4.937	
17.53	7.18	24.30	.126	0.	0.00	41.60	3.545	
18.09	8.45	24.50	.161	0.	0.00	50.20	3.115	
19.23	7.20	24.10	.190	0.	0.00	63.80	2.312	

EXPERIMENT #2,FLASK C,AFTER 3RD DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
16.85	7.18	25.00	.091	0.	0.00	30.30	2.630	
17.06	7.10	26.00	.094	0.	0.00	30.30	4.509	
17.53	7.13	26.00	.136	0.	0.00	44.40	3.364	
18.09	7.17	26.00	.179	0.	0.00	57.80	2.545	
19.23	7.25	26.00	.201	0.	0.00	66.20	2.272	

EXPERIMENT #2,FLASK L,AFTER 3RD DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
16.85	7.20	24.80	.093	0.	0.00	30.40	2.679	AFTER 50-50 DILUTION WITH FRESH MEDIUM
17.06	7.10	25.60	.096	0.	0.00	31.70	0.000	
17.53	7.18	25.20	.140	0.	0.00	46.80	3.316	
18.09	7.05	26.00	.181	0.	0.00	57.20	2.790	
19.23	7.20	25.80	.203	0.	0.00	68.40	2.173	

Table A-2. Continued.

EXPERIMENT #2,FLASK R,AFTER 4TH DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
19.26	7.45	23.90	.101	0.	0.00	33.00	2.187	AFTER 50-50 DILUTION, NO NITROGEN
20.34	7.20	24.00	.122	0.	0.00	40.00	2.159	
22.14	8.00	24.00	.136	0.	0.00	30.30	2.100	CO2 CYLINDER EXHAUSTED

EXPERIMENT #2,FLASK C,AFTER 4TH DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
19.26	7.37	25.00	.108	0.	0.00	36.00	1.944	AFTER 50-50 DILUTION, NO NITROGEN
20.34	7.12	25.60	.122	0.	0.00	38.00	2.016	
22.14	7.98	25.50	.132	0.	0.00	41.80	1.548	CO2 CYLINDER EXHAUSTED

EXPERIMENT #2,FLASK L,AFTER 4TH DILUTION

TIME (DAYS)	PH	TEMP DEG C	OD	CELLS/ML	MEAN CELL VOLUME (MICRON**3)	DRY WEIGHT (MG/L)	PERCENT NITROGEN	COMMENTS
19.26	7.40	25.80	.108	0.	0.00	34.70	2.251	AFTER 50-50 DILUTION, NO NITROGEN
20.34	7.05	25.50	.123	0.	0.00	0.00	0.000	
22.14	7.80	25.80	.138	0.	0.00	43.20	1.708	CO2 CYLINDER EXHAUSTED

Appendix B

Computer Program Listing

Table B-1.   Computer program listing.

```
                                                                     START OF SEGMENT 002
                                                                 FORMAT SEGMENT IS 00C9 LONG
        INTEGER TSTEP                                            C    002:000C:0
        REAL KN(4,4) ,M(4,1000),K1,K2,NA                        C    002:000C:0
        DIMENSION HEAD1(12),HEAD2(12),XLABEL(12),YLABL1(12),YLABL2(12)   C    002:000C:0
        DIMENSION Y(4),DATA(180,4),MHEAD(15),D(10,9),COON(200),C(7)      C    002:000C:0
        DIMENSION DILT(10)                                      C    002:000C:0
     DIMENSION A1(1045),Z1(180),Z2(500),Y1(180),Y2(500),X1(180),X2(500) C    002:000C:0
        DIMENSION YB(200)                                       C    002:000C:0
        DIMENSION YLABL3(12),N1(180),N2(500),HEAD3(12)          C    002:000C:0
        DATA A1/1045*'        '/                                C    002:000C:0
        READ (5,924)IEQOPT,IDILT,IPLOT,(C(L),L=1,7),TSTEP       C    002:000C:0
                                                                      FIB IS C006 LONG
  924   FORMAT (2I2,I1,7F10.0,I31                              C    002:0014:2
        IF(IEQOPT.EQ.2) IEQ=2                                  C    002:0014:2
        IF (IDILT.EQ.0) GO TO 253                              C    002:0016:3
        DO 253 ITIC=1,IDILT                                   C    002:0017:4
        READ (5,928) DILT(ITIC),(D(ITIC,L) ,L=1,9)            C    002:0019:0
  253   CONTINUE                                               C    002:002A:2
  928   FORMAT (10F5.0)                                        C    002:002C:3
  242   READ (5,904) HEAD1,YLABL1,XLABEL,HEAD2,YLABL2,YLABL3,HEAD3   C    002:002C:3
  904   FORMAT (12A6,8X)                                       C    002:003E:2
        READ (5,9111) COON                                     C    002:003E:2
 9111   FORMAT (10A6,20X)                                      C    002:0045:2
        READ(5,921) LQ,MHEAD                                   C    002:0045:2
  921   FORMAT (I3,15A2)                                       C    002:004E:2
        READ (5,922) TBAR,TMIN,TMAX,PHBAR,PHMIN,PHMAX          C    002:004E:2
  922   FORMAT (6E9.3)                                         C    002:005F:2
        DO 241 K=1,LQ                                          C    002:005F:2
  241   READ (5,923)(DATA(K,L),L=1,4)                          C    002:006C:0
  923   FORMAT (4F10.5)                                        C    002:007C:3
        ITIC=1                                                 C    002:007C:3
        DOGG=C(6)                                              C    002:0071:1
        DDGC=C(6)                                              C    002:0072:2
        DO 7821 IET=1,LQ                                       C    002:0073:3
        IF (IDILT.EQ.0)GO TO 7821                              C    002:0075:0
        IF (DATA(IET,1).LT.DILT(ITIC))GO TO 7821              C    002:0076:1
        DDGC=D(ITIC,9)                                         C    002:0078:5
        ITIC=ITIC+1                                            C    002:007A:3
        IF (ITIC.GT.IDILT)ITIC=IDILT                          C    002:0078:5
 7821   DATA(IET,4)=DDGC*DATA(IET,4)                           C    002:007D:4
        WRITE (6,9141) TBAR,TMIN,TMA),MHEAD,PHBAR,PHMIN,PHNA),C(6)   C    002:0083:2
                                                                      FIB IS 0006 LONG
 9141   FORMAT (1H1,T32,'OBSERVED EXPERIMENTAL DATA*,T67,'MEAN TEMP=',   C    002:0095:2
       1 F4.1,T86,'RANGE',F5.1,' TO',F5.1/T32,15A2,T67,'MEAN PH',F4.1,   C    002:0095:2
       1 T86,'RANGE',F5.1,' TC',F5.1/T86,'TOTAL NITROGEN=',F5.2)        C    002:0095:2
        WRITE (6,9142)                                         C    002:0095:2
 9142   FORMAT (1H ,T59,'DRY',T65,'CELLULAR', T75,'EXTERNAL'/T50,'TIME', C    002:0099:2
       1 T59,'WT.',T69,'N',T79,'N'/T45,'(DATS)',T58,'(MG/L)',T67,'(PG/L)', C    002:0099:2
       1 T76,'(MG/L)')                                         C    002:0099:2
        DO 7142 L=1,LQ                                         C    002:0099:2
 7142   WRITE (6,9143) (DATA(L,K),K=1,4)                       C    002:009A:0
 9143   FORMAT (1H ,T49,4(F5.2,5))                             C    002:00A9:3
        T=0.                                                   C    002:00A9:3
C       LRUN=LENGTH OF RUN IN HOURS                            C    002:00AA:1
C       DEFINE INTIAL CONDITIONS                               C    002:00AA:1
        N(1,1)=DATA(1,4)                                       C    002:00AA:1
        N(2,1)=DATA(1,2)                                       C    002:00AB:5
        PERN2=9999.                                            C    002:00AD:2
```

Table B-1. Continued.

```
C        DEFINE TIME STEP & RELATED PARAMETERS              C   002:00AE:2
         LRUN=(DATA(L0,1)-DATA(1,1))*24.+1                  C   002:00AE:2
C        TSTEP=TIME STEP IN MINUTES(INTEGER)                C   002:00B1:3
C LO IS THE NUMBER OF TIME STEPS IN A HOUR                  C   002:00B1:3
         L0=60/TSTEP                                        C   002:00B1:3
C        NUM=NUMBER OF TIME STEPS                           C   002:00B3:0
         NUM=LRUN*60/TSTEP+1                                C   002:00B3:0
         IF (NUM-1000)98,98,99                              C   002:00B5:2
  99     WRITE (6,901)                                      C   002:00B6:3
 901     FORMAT (1H ,T30,'NUMBER OF TIME STEPS NOT COMPATIBLE WITH    C   002:008A:2
        1 DIMENSIONED ARRAYS')                              C   002:008A:2
         GO TO 899                                          C   002:008A:2
  98     CONTINUE                                           C   002:009A:5
C        DELTA=TIME STEP IN DAYS                            C   002:008A:5
         DELTA=TSTEP/1440.                                  C   002:008A:5
         WRITE (6,9112) COON                                C   002:009C:2
 9112    FORMAT (1H1,T35,10A6/9(T5,10A6,5X,10A6/))          C   002:00C3:2
         C(6)=DCGG                                          C   002:00C3:2
         WRITE (6,9113) (K,C(K),K=1,7)                      C   002:00C4:4
 9113    FORMAT (1H ,T54,'DEFINITION OF CONSTANTS'/T2,7('C(',I1,')=',  C   0C2:00D2:2
        1 E9.3,4X)/T50,'DEFINITION OF DILUTION CONSTANTS')  C   002:00D2:2
         DO 8819 ITIC=1,IDILT                               C   002:00D2:2
 8819    WRITE (6,9003) DILT(ITIC),(L,D(ITIC,L),L=1,9)      C   002:00D3:0
 9003    FORMAT (1H ,T54,'TIME OF DILUTION=',F5.2/T2,9('D(',I1,')=',F6.3,3  C   002:00E6:3
        1X))                                                C   002:00E6:3
         WRITE (6,9131) (COON(K),K=1,10),TSTEP              C   002:00E6:3
 9131    FORMAT (1H1,T40,10A6/T53,'RESULTS OF ALGAL GROWTH SIMULATION'/  C   002:00F4:2
        1T53,'NUMBER OF MINUTES IN A TIME STEP IS',I2)      C   002:00F4:2
         WRITE (6,9135)                                     C   002:00F4:2
 9135    FORMAT (1H0,T50,'TIME',T62,'N',T77,'X',T88,'PERCENT N')  C   002:00F8:2
         PERN1=(C(6)-NC1,1))/N(2,1)*100.                    C   002:00F8:2
         WRITE (6,9136) DATA(1,1),(N(J,1),J=1,2),PERN1      C   0C2:00F8:2
 9136    FORMAT (1H ,T49,F5.2,T56,E12.5,T71,E12.5,T86,E12.5)  C   002:010A:2
         L=0                                                C   002:010A:2
         LF=0                                               C   002:010B:0
         ILINE=7                                            C   002:010B:4
         IV=0                                               C   002:010C:3
         ITIC=1                                             C   002:010D:1
         LDILT=(DILT(ITIC)-DATA(1,1))/DELTA                 C   002:010D:5
         DO 100 I=2,NUM                                     C   002:011C:2
         IF (IDILT.EQ.0) GO TO 7777                         C   002:0112:0
         IV=IV+1                                            C   002:0113:1
         IF (IV.NE.LDILT) GO TO 7777                        C   002:0114:3
         N(1,I-1)=D(ITIC,1)*N(1,I-1)+D(ITIC,2)              C   002:0115:3
         N(2,I-1)=D(ITIC,3)*N(2,I-1)+D(ITIC,4)              C   002:011A:2
         N(3,I-1)=D(ITIC,5)*N(3,I-1)+D(ITIC,6)              C   002:011F:2
         N(4,I-1)=D(ITIC,7)*N(4,I-1)+D(ITIC,8)              C   002:0124:2
         C(6)=D(ITIC,9)                                     C   002:0129:2
         PERN2=9999.                                        C   002:012B:3
         ITIC=ITIC+1                                        C   002:012C:3
         LDILT=(DILT(ITIC)-DATA(1,1))/DELTA                 C   002:012D:5
 7777    CONTINUE                                           C   002:013C:2
         LF=LF+1                                            C   002:013C:2
C        DEFINE K1'S FOR FOURTH ORDER RUNGE KUTTA           C   002:0131:4
         DO 123 J=1,IEQ                                     C   002:0131:4
 123     Y(J)=N(J,I-1)                                      C   002:0133:0
         J=1                                                C   002:0138:4
         GO TO 600                                          C   002:0139:2
C        DEFINE K2'S FOR FOURTH ORDER RUNGE KUTTA           C   0C2:0139:5
 125     CONTINUE                                           C   002:0139:5
         DO 128 J=1,IEQ                                     C   002:0139:5
```

Table B-1. Continued.

```
      128  Y(J)=N(J,I-1)+DELTA/3*KN(J,1)               C   002:0138:0
           J=2                                         C   002:0142:5
           GO TO 600                                   C   002:0143:4
      C         DEFINE X3'S FOR FOURTH ORDER RUNGE KUTTA    C   002:0144:1
      150  CONTINUE                                    C   002:0144:1
           DO 158 J=1,IEQ                              C   002:0144:1
      158  Y(J)=N(J,I-1)-DELTA/3*KN(J,1)+DELTA*KN(J,2)  C   002:0145:0
           J=3                                         C   002:014E:4
           GO TO 600                                   C   002:014F:3
      C         DEFINE X4'S FOR FOURTH ORDER RUNGE KUTTA    C   002:015C:0
      175  CONTINUE                                    C   002:015C:0
           DO 178 J=1,IEQ                              C   002:015C:0
      178  Y(J)=N(J,I-1)+DELTA*(KN(J,1)-KN(J,2)+KN(J,3))  C   002:0151:0
           J=4                                         C   002:015B:0
           GO TO 600                                   C   002:015B:5
      C         DEFINE NEW NUTRIENT LEVEL BY APPLICATION OF RUNGE KUTTA   C   002:015C:2
      200  CONTINUE                                    C   002:015C:2
           DO 222 J=1,IEQ                              C   002:015C:2
      222  N(J,I)=N(J,I-1)+DELTA/6.*(KN(J,1)+2*KN(J,2)+2*KN(J,3)+KN(J,4))  C   002:015D:0
           W2(I)=(C(6)-N(1,I))/N(2,I)*100.            C   002:016B:0
           IF (LF-LD)228,227,227                       C   002:0171:1
      227  L=L+1                                       C   002:0172:3
           LF=0                                        C   002:0173:5
           IF (ILINE.LT.60) GO TO 7312                 C   002:0174:3
           ILINE=6                                     C   002:0175:5
           WRITE (6,9131) (COON(K),K=1,10),TSTEP       C   002:0176:4
           WRITE (6,9135)                              C   002:0184:2
      7312 CONTINUE                                    C   002:0188:2
           PERN3=PERN2                                 C   002:0188:2
           TR=DATA(1,1)+I*DELTA                        C   002:0189:1
           PERN2=(C(6)-N(1,I))/N(2,I)*100.            C   002:018B:1
           WRITE (6,9136) TR,(N(J,I),J=1,2),PERN2      C   002:0190:3
           ILINE=ILINE+1                               C   002:01AC:2
           IPNT=1                                      C   002:01A1:4
           IF(PERN2-PERN3.GT.0.)IPNT=2                 C   002:01A2:2
           IF (PERN2.GE.C(4))IPNT=1                    C   002:01A4:5
           IF(PERN2-C(7).LT..001)IPNT=3               C   002:01A7:1
      228  CONTINUE                                    C   002:01A8:3
      100  CONTINUE                                    C   002:01A8:3
           GO TO 899                                   C   002:01A0:4
      600  CONTINUE                                    C   002:01AE:1
           PERN1=(C(6)-Y(1))/Y(2)*100.-C(7)           C   002:01AE:1
           KN(1,J)=-C(1)*Y(1)*Y(2)                    C   002:01B2:0
           GO TO (8814,8815,8816)IPNT                  C   002:01B6:0
      8814 KN(2,J)=C(2)*PERN1*Y(2)                    C   002:01BB:3
           GO TO 88                                    C   002:01BF:1
      8815 KN(2,J)=C(3)*Y(2)                          C   002:018F:4
           GO TO 88                                    C   002:01C3:0
      8816 KN(2,J)=0.0                                 C   002:01C3:3
       88  CONTINUE                                    C   002:01C5:4
           GO TO (125,150,175,200) .                  C   002:01C5:4
      899  CONTINUE                                    C   002:01CB:3
      882  LN=1                                        C   002:01CB:3
           LX=2                                        C   002:01CC:1
           IF (IPLOT.NE.1) GO TO 891                   C   002:01C0:0
           XMAXT=DATA(1,1)+12.                         C   002:01CE:1
           XMIN=DATA(1,1)                              C   002:01CF:4
           XMAX=DATA(1,1)+(NUM-1)*DELTA               C   002:01D0:4
           IF (XMAX.LT.XMAXT) XMAX=XMAXT              C   002:01D3:0
           WMIN=0.                                     C   002:01D4:5
           WMAX=10.                                    C   002:01D5:3
```

Table B-1. Continued.

```
        YMIN=0.                                              C   002:0106:2
        ZMIN=0.                                              C   002:0107:0
        ZMAX=100.                                            C   002:0107:4
        YMAX=1.0                                             C   002:0108:3
892     DO 894 I=1,LG                                        C   002:0109:1
        W1(I)=DATA(I,3)/DATA(I,2)*100.                       C   002:010A:0
        IF (YMIN.GT.DATA(I,4))    YMIN=DATA(I,4)             C   002:010E:3
        IF (ZMIN.GT.DATA(I,2))    ZMIN=DATA(I,2)             C   002:01E2:4
        IF (WMIN.GT.W1(I)) WMIN=W1(I)                        C   002:01E6:3
        IF (WMAX.LT.W1(I)) WMAX=W1(I)                        C   002:01EA:0
        IF (YMAX.LT.DATA(I,4))    YMAX=DATA(I,4)             C   002:01ED:3
        IF (ZMAX.LT.DATA(I,2))    ZMAX=DATA(I,2)             C   002:01F1:4
        YI(I)=DATA(I,4)                                      C   002:01F5:3
        Z1(I)=DATA(I,2)                                      C.  002:01F8:1
        X1(I)=DATA(I,1)                                      C   002:01FA:4
894     CONTINUE                                             C   002:01FD:0
        DO 895 I=1,NUM                                       C   002:01FF:1
        IF (WMAX.LT.W2(I)) YMAX=W2(I)                        C   002:0200:0
        IF (WMIN.GT.W2(I)) WMIN=W2(I)                        C   002:0203:3
        IF (YMIN.GT.N(LN,I))      YMIN=N(LN,I)               C   002:0207:0
        IF (YMAX.LT.N(LN,I))      YMAX=N(LN,I)               C   002:020C:5
        IF (ZMIN.GT.N(LX,I))      ZMIN=N(LX,I)               C   002:0212:4
        IF (ZMAX.LT.N(LX,I))      ZMAX=N(LX,I)               C   002:0218:3
        Y2(I)=N(LN,I)                                        C   002:021E:2
        Z2(I)=N(LX,I)                                        C   002:0221:5
        X2(I)=DATA(1,1)+(I-1)*DELTA                          C   002:0225:2
895     CONTINUE                                             C   002:0228:3
        CALL PL360   (NUM,A1,X2,XMIN,XMAX,XLABEL,Y2,YMIN,YMAX,YLABL1,HEAD1,   C   002:022A:4
       1-240)                                                C   002:0231:4
        CALL PL360   (LQ,A1,X1,XMIN,XMAX,XLABEL,YI,YMIN,YMAX,YLABL1,HEAD1,    C   002:0232:2
       1213)                                                 C   002:0239:2
        WRITE (6,9144)                                       C   002:0239:5
        WRITE (6,9147) C(7),C(1),C(2)                        C   002:023E:2
9144    FORMAT (1H ,T14,'OBSERVED CATA INDICATED BY "N"'/T14,'SIMULATED C     C   002:0246:2
       1URVE COMPOSED OF "G"')                               C   002:0246:2
        CALL PL360   (NUM,A1,X2,XMIN,XMAX,XLABEL,Z2,ZMIN,ZMAX,YLABL2,HEAD2,   C   002:0248:2
       1-240)                                                C   002:024F:2
        CALL PL360   (LQ,A1,X1,XMIN,XMAX,XLABEL,Z1,ZMIN,ZMAX,YLABL2,HEAD2,    C   002:025C:0
       1231)                                                 C   002:0257:0
        WRITE (6,9145)                                       C   002:0257:3
        WRITE (6,9147) C(7),C(1),C(2)                        C   002:0258:2
9145    FORMAT (1H ,T14,'OBSERVED CATA INDICATED BY "X"'/T14,'SIMULATED C     C   002:0265:2
       1URVE COMPOSED OF "D"')                               C   002:0265:2
        CALL PL360   (NUM,A1,X2,XMIN,XMAX,XLABEL,W2,WMIN,WMAX,YLABL3,         C   002:0265:2
       * HEAD3,-240)                                         C   002:0268:4
        CALL PL360 (LQ,A1,X1,XMIN,XM/X,XLABEL,W1,WMIN,WMAX,YLABL3,            C   002:026D:0
       * HEAD3,215)                                          C   002:0273:2
        WRITE (6,9146)                                       C   002:0274:3
9146    FORMAT (1H ,T14,'OBSERVEC CATA INDICATED BY "P"'/T14,'SIMULATED C     C   002:0278:2
       *URVE COMPOSED OF "D"')                               C   002:0278:2
        WRITE (6,9147) C(7),C(1),C(2)                        C   002:0278:2
9147    FORMAT (1H ,T14,'CONSTANTS FCR THIS RUN:WMIN='F4.2,'JG=',F4.2,        C   002:0282:2
       * 'JU=',F4.2)                                         C   002:0282:2
891     CONTINUE                                             C   002:0282:2
        CALL EXIT                                            C   002:0282:2
        END                                                  C   002:0283:1
```

SEGMENT 002 IS 0283 LONG

Appendix C

Output Listings for Simulation Runs

Table C-1. Simulation of experiment #1. (See Figures 10 - 12 in text.)

OBSERVED EXPERMENTAL DATA   MEAN TEMP=25.0   RANGE 23.4 TO 27.5
MEAN VALUES OF RUNS 1+2   MEAN PH 7.1   RANGE 6.5 TO 10.0
                                                              TOTAL NITROGEN= 1.60

	DRY	CELLULAR	EXTERNAL
TIME	WT.	N	N
(DAYS)	(MG/L)	(MG/L)	(MG/L)
1.83	26.23	1.33	0.27
2.10	36.54	1.41	0.19
2.67	51.20	1.30	0.30
2.85	51.60	1.44	0.16
3.16	57.67	1.45	0.15
3.55	61.40	1.60	0.00
4.18	71.60	1.60	-0.00
4.70	72.30	1.76	-0.16
4.73	36.40	0.76	0.84
4.88	36.60	1.02	0.71
5.05	37.07	1.50	0.23
5.20	41.00	1.57	0.16
5.52	49.00	1.64	0.09
5.81	55.20	1.73	-0.00

Table C-1. Continued.

```
DN/DT=-G*N*X     DX/DT=U*(PERN-NMIN)*X                    DILUTION ROUTINE (OPTIONAL)
N=EXTRACELLULAR NITROGEN (MG/L)                           N=D(1)*N+D(2)      X=D(3)*X+D(4)
X=ALGAL BIOMASS AS DRY WT. (MG/L)                         C(6)=D(9)
PERN=PERCENT NITROGEN=AT(T-N)/X
NMIN=MINIMUM PERCENT NITROGEN FOR POSITIVE GROWTH
NTOT=TOTAL NITROGEN (MG/L)                                     <
C(1)=G   C(2)=U   C(6)=NTOT    C(7)=NMIN
```

```
                                     DEFINITION OF CONSTANTS
C(1)= .100E+00     C(2)= .390E+00     C(3)= .500E-01    C(4)= .380E+01    C(5)=0.           C(6)= .160E+01    C(7)= .211E+01
                                     DEFINITION OF DILUTION CONSTANTS
                                     TIME OF DILUTION= 4.73
D(1)= 0.000    D(2)= 0.930    D(3)= 0.500    D(4)= 0.000    D(5)= 0.000    D(6)= 0.000    D(7)= 0.000    D(8)= 0.000    D(9)= 1.730
```

Table C-1. Continued.

EQUATION SET #3
RESULTS OF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS 30

TIME	N	X	PERCENT N
1.83	.27426E+00	.26233E+02	.50536E+01
1.90	.24522E+00	.27490E+02	.49282E+01
1.94	.21811E+00	.28750E+02	.48066E+01
1.98	.19298E+00	.30009E+02	.46887E+01
2.02	.16985E+00	.31264E+02	.45744E+01
2.06	.14872E+00	.32512E+02	.44638E+01
2.10	.12954E+00	.33750E+02	.43569E+01
2.15	.11226E+00	.34976E+02	.42536E+01
2.19	.96794E-01	.36186E+02	.41541E+01
2.23	.83040E-01	.37379E+02	.40583E+01
2.27	.70890E-01	.38552E+02	.39663E+01
2.31	.60225E-01	.39704E+02	.38781E+01
2.35	.50922E-01	.40833E+02	.37937E+01
2.40	.42856E-01	.41938E+02	.37129E+01
2.44	.35904E-01	.43018E+02	.36359E+01
2.48	.29947E-01	.44071E+02	.35625E+01
2.52	.24870E-01	.45098E+02	.34927E+01
2.56	.20566E-01	.46098E+02	.34263E+01
2.60	.16938E-01	.47070E+02	.33632E+01
2.65	.13894E-01	.48015E+02	.33034E+01
2.69	.11353E-01	.48932E+02	.32466E+01
2.73	.92417E-02	.49823E+02	.31928E+01
2.77	.74957E-02	.50686E+02	.31419E+01
2.81	.60581E-02	.51523E+02	.30937E+01
2.85	.48794E-02	.52333E+02	.30480E+01
2.90	.39171E-02	.53118E+02	.30048E+01
2.94	.31344E-02	.53878E+02	.29639E+01
2.98	.25003E-02	.54613E+02	.29251E+01
3.02	.19885E-02	.55325E+02	.28884E+01
3.06	.15768E-02	.56013E+02	.28536E+01
3.10	.12469E-02	.56679E+02	.28207E+01
3.15	.98328E-03	.57323E+02	.27895E+01
3.19	.77337E-03	.57945E+02	.27599E+01
3.23	.60672E-03	.58547E+02	.27318E+01
3.27	.47481E-03	.59129E+02	.27052E+01
3.31	.37069E-03	.59691E+02	.26799E+01
3.35	.28874E-03	.60234E+02	.26558E+01
3.40	.22441E-03	.60759E+02	.26330E+01
3.44	.17403E-03	.61267E+02	.26112E+01
3.48	.13469E-03	.61758E+02	.25906E+01
3.52	.10403E-03	.62232E+02	.25709E+01
3.56	.80190E-04	.62690E+02	.25521E+01
3.60	.61700E-04	.63133E+02	.25342E+01
3.65	.47387E-04	.63561E+02	.25172E+01
3.69	.36330E-04	.63974E+02	.25010E+01
3.73	.27807E-04	.64374E+02	.24854E+01
3.77	.21248E-04	.64760E+02	.24706E+01
3.81	.16210E-04	.65133E+02	.24565E+01
3.85	.12348E-04	.65493E+02	.24430E+01
3.90	.93925E-05	.65842E+02	.24301E+01
3.94	.71341E-05	.66178E+02	.24177E+01
3.98	.54112E-05	.66504E+02	.24059E+01
4.02	.40990E-05	.66818E+02	.23946E+01
4.06	.31009E-05	.67122E+02	.23837E+01

Table C-1. Continued.

EQUATION SET #3
RESULTS OF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS30

TIME	N	X	PERCENT N
4.10	.23430E-05	.67415E+02	.23733E+01
4.15	.17682E-05	.67699E+02	.23634E+01
4.19	.13328E-05	.67973E+02	.23539E+01
4.23	.10036E-05	.68238E+02	.23447E+01
4.27	.75481E-06	.68494E+02	.23360E+01
4.31	.56712E-06	.68741E+02	.23276E+01
4.35	.42567E-06	.68980E+02	.23195E+01
4.40	.31919E-06	.69211E+02	.23118E+01
4.44	.23912E-06	.69434E+02	.23044E+01
4.48	.17897E-06	.69649E+02	.22972E+01
4.52	.13383E-06	.69858E+02	.22904E+01
4.56	.99994E-07	.70059E+02	.22838E+01
4.60	.74650E-07	.70253E+02	.22775E+01
4.65	.55685E-07	.70441E+02	.22714E+01
4.69	.41506E-07	.70623E+02	.22656E+01
4.73	.30915E-07	.70798E+02	.22599E+01
4.77	.80220E+00	.35589E+02	.26070E+01
4.81	.69115E+00	.35963E+02	.28887E+01
4.85	.59488E+00	.36038E+02	.31498E+01
4.90	.51186E+00	.36113E+02	.33731E+01
4.94	.44028E+00	.36188E+02	.35639E+01
4.98	.37860E+00	.36264E+02	.37266E+01
5.02	.32545E+00	.36339E+02	.38651E+01
5.06	.27911E+00	.37396E+02	.38798E+01
5.10	.23830E+00	.38487E+02	.38758E+01
5.15	.20253E+00	.39602E+02	.38571E+01
5.19	.17132E+00	.40733E+02	.38266E+01
5.23	.14423E+00	.41872E+02	.37872E+01
5.27	.12085E+00	.43013E+02	.37410E+01
5.31	.10079E+00	.44151E+02	.36901E+01
5.35	.83654E-01	.45279E+02	.36360E+01
5.40	.69111E-01	.46395E+02	.35799E+01
5.44	.56832E-01	.47494E+02	.35229E+01
5.48	.46524E-01	.48575E+02	.34657E+01
5.52	.37915E-01	.49634E+02	.34091E+01
5.56	.30765E-01	.50670E+02	.33535E+01
5.60	.24857E-01	.51681E+02	.32993E+01
5.65	.20001E-01	.52667E+02	.32468E+01
5.69	.16028E-01	.53627E+02	.31961E+01
5.73	.12793E-01	.54560E+02	.31474E+01
5.77	.10173E-01	.55467E+02	.31007E+01
5.81	.80588E-02	.56346E+02	.30560E+01
5.85	.63612E-02	.57199E+02	.30134E+01

Table C-2. Simulation of experiment #2 before dilution. (See Figures 13 - 15 in text.)

OBSERVED EXPERMENTAL DATA   MEAN TEMP=24.9   RANGE 22.C TO 26.0
MEAN VALUES OF RUNS 4 THRU 6   MEAN PH 7.1   RANGE 6.9 TO 9.5
                                                          TOTAL NITROGEN= 1.68

TIME (DAYS)	DRY WT. (MG/L)	CELLULAR N (MG/L)	EXTERNAL N (MG/L)
11.06	38.93	0.90	0.78
11.52	38.60	1.26	0.42
14.97	77.67	1.71	-0.03
15.64	80.00	1.73	-0.05
15.67	38.73	0.75	0.74
16.05	40.93	1.35	0.18
16.56	55.47	1.55	-0.02
16.83	59.40	1.53	-0.00
16.85	30.23	0.82	0.71
17.06	30.77	1.43	0.10
17.53	44.27	1.51	0.02
18.09	55.07	1.54	-0.01
19.23	66.13	1.45	0.04

Table C-2. Continued.

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DN/DT=-G*N*X     DX/DT=U*(PERN-NMIN)*X                        DILUTION ROUTINE (OPTIONAL)
N=EXTRACELLULAR NITROGEN (MG/L)                               N=D(1)*N+D(2)     X=D(3)*X+D(4)
X=ALGAL BIOMASS AS DRY WT. (MG/L)                            C(6)=D(9)
PERN=PERCENT NITROGEN=NTOT-N)/X
NMIN=MINIMUM PERCENT NITROGEN FOR POSITIVE GROWTH
NTOT=TOTAL NITROGEN (MG/L)                                    C(1)=G    C(2)=U    C(6)=NTOT    C(7)=NMIN
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                                        DEFINITION OF CONSTANTS
C(1)= .100E+00    C(2)= .390E+0C    C(3)= .500E-01    C(4)= .300E+01    C(5)=0.            C(6)= .168E+01    C(7)= .211E+01
                                        DEFINITION OF DILUTION CONSTANTS
                                          TIME OF DILUTION=15.67
D(1)= 0.000    D(2)= 0.665    D(3)= 0.500    D(4)= 0.000    D(5)= 0.000    D(6)= 0.000    D(7)= 0.000    D(8)= 0.000    D(9)= 1.530
                                          TIME OF DILUTION=16.85
D(1)= 0.000    D(2)= 0.665    D(3)= 0.5C0    D(4)= 0.000    D(5)= 0.000    D(6)= 0.000    D(7)= 0.000    D(8)= 0.000    D(9)= 1.530
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Table C-2.   Continued.

EQUATION SET #3
RESULTS OF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS 30

TIME	N	X	PERCENT N
11.06	.77964E+00	.38933E+02	.23126E+01
11.12	.66263E+00	.39156E+02	.25983E+01
11.16	.56245E+00	.39544E+02	.28261E+01
11.21	.47693E+00	.39626E+02	.30360E+01
11.25	.40381E+00	.40273E+02	.31689E+01
11.29	.34092E+00	.41006E+02	.32656E+01
11.33	.28691E+00	.41807E+02	.33322E+01
11.37	.24062E+00	.42661E+02	.33740E+01
11.41	.20106E+00	.43555E+02	.33955E+01
11.46	.16737E+00	.44478E+02	.34009E+01
11.50	.13879E+00	.45418E+02	.33934E+01
11.54	.11463E+00	.46370E+02	.33759E+01
11.58	.94305E-01	.47324E+02	.33507E+01
11.62	.77275E-01	.48276E+02	.33199E+01
11.66	.63071E-01	.49221E+02	.32850E+01
11.71	.51276E-01	.50155E+02	.32474E+01
11.75	.41527E-01	.51074E+02	.32080E+01
11.79	.33503E-01	.51976E+02	.31678E+01
11.83	.26930E-01	.52860E+02	.31272E+01
11.87	.21568E-01	.53724E+02	.30870E+01
11.91	.17212E-01	.54566E+02	.30473E+01
11.96	.13688E-01	.55386E+02	.30086E+01
12.00	.10849E-01	.56183E+02	.29709E+01
12.04	.85710E-02	.56958E+02	.29345E+01
12.08	.67497E-02	.57709E+02	.28994E+01
12.12	.52990E-02	.58438E+02	.28658E+01
12.16	.41477E-02	.59145E+02	.28335E+01
12.21	.32372E-02	.59829E+02	.28026E+01
12.25	.25194E-02	.60492E+02	.27731E+01
12.29	.19555E-02	.61133E+02	.27449E+01
12.33	.15138E-02	.61753E+02	.27181E+01
12.37	.11689E-02	.62353E+02	.26924E+01
12.41	.90041E-03	.62934E+02	.26680E+01
12.46	.69191E-03	.63495E+02	.26448E+01
12.50	.53048E-03	.64038E+02	.26226E+01
12.54	.40580E-03	.64562E+02	.26015E+01
12.58	.30976E-03	.65069E+02	.25814E+01
12.62	.23596E-03	.65559E+02	.25622E+01
12.66	.17938E-03	.66033E+02	.25439E+01
12.71	.13611E-03	.66491E+02	.25265E+01
12.75	.10308E-03	.66933E+02	.25098E+01
12.79	.77924E-04	.67360E+02	.24939E+01
12.83	.58804E-04	.67774E+02	.24788E+01
12.87	.44301E-04	.68173E+02	.24643E+01
12.91	.33320E-04	.68559E+02	.24504E+01
12.96	.25021E-04	.68931E+02	.24372E+01
13.00	.18761E-04	.69292E+02	.24245E+01
13.04	.14046E-04	.69640E+02	.24124E+01
13.08	.10501E-04	.69976E+02	.24008E+01
13.12	.78402E-05	.70301E+02	.23897E+01
13.16	.58457E-05	.70615E+02	.23791E+01
13.21	.43529E-05	.70919E+02	.23689E+01
13.25	.32374E-05	.71212E+02	.23591E+01
13.29	.24048E-05	.71496E+02	.23498E+01

Table C-2.   Continued.

EQUATION SET #3
RESULTS CF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS30

TIME	N	X	PERCENT N
13.33	.17843E-05	.71770E+02	.23408E+01
13.37	.13224E-05	.72034E+02	.23322E+01
13.41	.97900E-06	.72290E+02	.23240E+01
13.46	.72403E-06	.72537E+02	.23161E+01
13.50	.53492E-06	.72776E+02	.23085E+01
13.54	.39481E-06	.73007E+02	.23012E+01
13.58	.29113E-06	.73229E+02	.22942E+01
13.62	.21448E-06	.73445E+02	.22874E+01
13.66	.15787E-06	.73653E+02	.22810E+01
13.71	.11610E-06	.73854E+02	.22748E+01
13.75	.85318E-07	.74049E+02	.22688E+01
13.79	.62644E-07	.74236E+02	.22630E+01
13.83	.45961E-07	.74418E+02	.22575E+01
13.87	.33696E-07	.74593E+02	.22522E+01
13.91	.24686E-07	.74763E+02	.22471E+01
13.96	.18073E-07	.74926E+02	.22422E+01
14.00	.13222E-07	.75085E+02	.22375E+01
14.04	.96673E-08	.75238E+02	.22329E+01
14.08	.70637E-08	.75385E+02	.22286E+01
14.12	.51582E-08	.75528E+02	.22243E+01
14.16	.37645E-08	.75666E+02	.22203E+01
14.21	.27459E-08	.75799E+02	.22164E+01
14.25	.20017E-08	.75928E+02	.22126E+01
14.29	.14585E-08	.76053E+02	.22090E+01
14.33	.10621E-08	.76173E+02	.22055E+01
14.37	.77313E-09	.76289E+02	.22021E+01
14.41	.56248E-09	.76401E+02	.21989E+01
14.46	.40904E-09	.76510E+02	.21958E+01
14.50	.29733E-09	.76615E+02	.21928E+01
14.54	.21603E-09	.76716E+02	.21899E+01
14.58	.15690E-09	.76814E+02	.21871E+01
14.62	.11390E-09	.76909E+02	.21844E+01
14.66	.82659E-10	.77000E+02	.21818E+01
14.71	.59963E-10	.77088E+02	.21793E+01
14.75	.43483E-10	.77174E+02	.21769E+01
14.79	.31522E-10	.77256E+02	.21746E+01
14.83	.22843E-10	.77336E+02	.21723E+01
14.87	.16548E-10	.77413E+02	.21702E+01
14.91	.11984E-10	.77487E+02	.21681E+01
14.96	.86762E-11	.77559E+02	.21661E+01
15.00	.62795E-11	.77629E+02	.21641E+01
15.04	.45436E-11	.77696E+02	.21623E+01
15.08	.32867E-11	.77761E+02	.21605E+01
15.12	.23768E-11	.77823E+02	.21587E+01
15.16	.17184E-11	.77884E+02	.21571E+01
15.21	.12421E-11	.77943E+02	.21554E+01
15.25	.89757E-12	.77999E+02	.21539E+01
15.29	.64847E-12	.78054E+02	.21524E+01
15.33	.46839E-12	.78107E+02	.21509E+01
15.37	.33825E-12	.78158E+02	.21495E+01
15.41	.24421E-12	.78207E+02	.21481E+01
15.46	.17629E-12	.78255E+02	.21468E+01
15.50	.12723E-12	.78301E+02	.21456E+01
15.54	.91803E-13	.78345E+02	.21444E+01

Table C-2.   Continued.

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                       EQUATION SET #3
                 RESULTS OF ALGAL GROWTH SIMULATION
                 NUMBER OF MINUTES IN A TIME STEP IS30

           TIME          N              X            PERCENT N
          15.58      .66231E-13      .78388E+02      .21432E+01
          15.62      .47773E-13      .78430E+02      .21420E+01
          15.66      .34454E-13      .78470E+02      .21409E+01
          15.71      .56457E+00      .39377E+02      .24517E+01
          15.75      .47888E+00      .39663E+02      .26501E+01
          15.79      .40586E+00      .39745E+02      .28283E+01
          15.83      .34386E+00      .39828E+02      .29781E+01
          15.87      .29123E+00      .39911E+02      .31038E+01
          15.91      .24627E+00      .40582E+02      .31633E+01
          15.96      .20766E+00      .41297E+02      .32021E+01
          16.00      .17456E+00      .42044E+02      .32238E+01
          16.04      .14628E+00      .42816E+02      .32318E+01
          16.08      .12218E+00      .43603E+02      .32287E+01
          16.12      .10171E+00      .44399E+02      .32169E+01
          16.16      .84392E-01      .45198E+02      .31984E+01
          16.21      .69790E-01      .45996E+02      .31746E+01
          16.25      .57523E-01      .46789E+02      .31471E+01
          16.29      .47257E-01      .47572E+02      .31168E+01
          16.33      .38698E-01      .48345E+02      .30847E+01
          16.37      .31588E-01      .49103E+02      .30516E+01
          16.41      .25703E-01      .49847E+02      .30178E+01
          16.46      .20851E-01      .50574E+02      .29841E+01
          16.50      .16864E-01      .51283E+02      .29506E+01
          16.54      .13600E-01      .51974E+02      .29176E+01
          16.58      .10937E-01      .52647E+02      .28854E+01
          16.62      .87706E-02      .53301E+02      .28540E+01
          16.66      .70146E-02      .53936E+02      .28237E+01
          16.71      .55956E-02      .54552E+02      .27944E+01
          16.75      .44524E-02      .55149E+02      .27662E+01
          16.79      .35340E-02      .55728E+02      .27391E+01
          16.83      .27985E-02      .56289E+02      .27131E+01
          16.87      .59083E+00      .28638E+02      .32794E+01
          16.91      .52375E+00      .29228E+02      .34428E+01
          16.96      .46306E+00      .29900E+02      .35684E+01
          17.00      .40820E+00      .30641E+02      .36611E+01
          17.04      .35868E+00      .31441E+02      .37255E+01
          17.08      .31409E+00      .32289E+02      .37657E+01
          17.12      .27405E+00      .33176E+02      .37858E+01
          17.16      .23822E+00      .34093E+02      .37890E+01
          17.21      .20627E+00      .35034E+02      .37785E+01
          17.25      .17790E+00      .35991E+02      .37568E+01
          17.29      .15282E+00      .36958E+02      .37263E+01
          17.33      .13074E+00      .37930E+02      .36890E+01
          17.37      .11140E+00      .38903E+02      .36465E+01
          17.41      .94543E-01      .39872E+02      .36002E+01
          17.46      .79912E-01      .40833E+02      .35513E+01
          17.50      .67276E-01      .41783E+02      .35007E+01
          17.54      .56416E-01      .42721E+02      .34493E+01
          17.58      .47126E-01      .43642E+02      .33978E+01
          17.62      .39217E-01      .44547E+02      .33466E+01
          17.66      .32513E-01      .45432E+02      .32961E+01
          17.71      .26857E-01      .46298E+02      .32467E+01
          17.75      .22107E-01      .47142E+02      .31986E+01
          17.79      .18133E-01      .47965E+02      .31520E+01
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Table C-2. Continued.

EQUATION SET #3
RESULTS CF ALGAL GROWTH SIMULATICN
NUMBER OF MINUTES IN A TIME STEP IS30

TIME	N	X	PERCENT N
17.83	.14823E-01	.48766E+02	.31070E+01
17.87	.12078E-01	.49545E+02	.30637E+01
17.91	.98097E-02	.50302E+02	.30221E+01
17.96	.79427E-02	.51037E+02	.29823E+01
18.00	.64116E-02	.51749E+02	.29442E+01
18.04	.51606E-02	.52440E+02	.29078E+01
18.08	.41419E-02	.53109E+02	.28731E+01
18.12	.33152E-02	.53757E+02	.28400E+01
18.16	.26465E-02	.54384E+02	.28084E+01
18.21	.21072E-02	.54992E+02	.27784E+01
18.25	.16737E-02	.55579E+02	.27498E+01
18.29	.13261E-02	.56147E+02	.27226E+01
18.33	.10483E-02	.56697E+02	.26967E+01
18.37	.82680E-03	.57229E+02	.26720E+01
18.41	.65070E-03	.57743E+02	.26486E+01
18.46	.51103E-03	.58240E+02	.26262E+01
18.50	.40052E-03	.58720E+02	.26049E+01
18.54	.31329E-03	.59184E+02	.25846E+01
18.58	.24459E-03	.59633E+02	.25653E+01
18.62	.19061E-03	.60067E+02	.25468E+01
18.66	.14828E-03	.60486E+02	.25293E+01
18.71	.11515E-03	.60891E+02	.25125E+01
18.75	.89273E-04	.61283E+02	.24965E+01
18.79	.69101E-04	.61661E+02	.24812E+01
18.83	.53404E-04	.62027E+02	.24666E+01
18.87	.41212E-04	.62380E+02	.24526E+01
18.91	.31757E-04	.62721E+02	.24393E+01
18.96	.24437E-04	.63051E+02	.24266E+01
19.00	.18778E-04	.63370E+02	.24144E+01
19.04	.14412E-04	.63678E+02	.24027E+01
19.08	.11046E-04	.63976E+02	.23915E+01
19.12	.84565E-05	.64264E+02	.23808E+01
19.16	.64663E-05	.64542E+02	.23705E+01
19.21	.49388E-05	.64810E+02	.23607E+01
19.25	.37680E-05	.65070E+02	.23513E+01
19.29	.28717E-05	.65321E+02	.23423E+01

Table C-3. Simulation of dilutions of experiment #2, transition point = 3.8 percent. (See Figures 16 - 18 in text.)

OBSERVED EXPERMENTAL DATA   MEAN TEMP=24.9   RANGE 22.0 TO 26.0
MEAN VALUES OF RUNS 4 THRU 6   MEAN PH 7.1   RANGE 6.9 TO 8.5
                                                      TOTAL NITROGEN= 1.68

TIME (DAYS)	DRY WT. (MG/L)	CELLULAR N (MG/L)	EXTERNAL N (MG/L)
11.06	38.93	0.90	0.78
11.52	38.60	1.26	0.42
14.97	77.67	1.71	-0.03
15.64	80.00	1.73	-0.05
15.67	38.73	0.75	0.74
16.05	40.93	1.35	0.18
16.56	55.47	1.55	-0.02
16.83	59.40	1.53	-0.00
16.85	30.23	0.82	0.71
17.06	30.77	1.43	0.10
17.53	44.27	1.51	0.02
18.09	55.07	1.54	-0.01
19.23	66.13	1.45	0.04

Table C-3. Continued.

EQUATION SET #3

DN/DT=-G*N*X   DX/DT=U*(PERN-NMIN)*X
N=EXTRACELLULAR NITROGEN (MG/L)
X=ALGAL BIOMASS AS DRY WT. (MG/L)
PERN=PERCENT NITROGEN=NTOT-N)/X
NMIN=MINIMUM PERCENT NITROGEN FOR POSITIVE GROWTH
NTOT=TOTAL NITROGEN (MG/L)

DILUTION ROUTINE (OPTIONAL)
N=D(1)*N+D(2)   X=D(3)*X+D(4)
C(6)=D(9)

C(1)=G   C(2)=U   C(6)=NTOT   C(7)=NMIN

DEFINITION OF CONSTANTS
C(1)= .100E+00   C(2)= .390E+00   C(3)= .500E-01   C(4)= .380E+01   C(5)=0.   C(6)= .168E+01   C(7)= .211E+01
DEFINITION OF DILUTION CONSTANTS
TIME OF DILUTION=15.67
D(1)= 0.000   D(2)= 0.665   D(3)= 0.500   D(4)= 0.000   D(5)= 0.000   D(6)= 0.000   D(7)= 0.000   D(8)= 0.000   D(9)= 1.530
TIME OF DILUTION=16.85
D(1)= 0.000   D(2)= 0.665   D(3)= 0.500   D(4)= 0.000   D(5)= 0.000   D(6)= 0.000   D(7)= 0.000   D(8)= 0.000   D(9)= 1.530

Table C-3. Continued.

EQUATION SET #3
RESULTS CF ALGAL GROWTH SIMULATICN
NUMBER OF MINUTES IN A TIME STEP IS30

TIME	N	X	PERCENT N
11.06	.77964E+00	.38933E+02	.23126E+01
11.12	.66263E+00	.39156E+02	.25983E+01
11.16	.56245E+00	.39544E+02	.28261E+01
11.21	.47693E+00	.39626E+02	.30360E+01
11.25	.40427E+00	.39709E+02	.32127E+01
11.29	.34257E+00	.39792E+02	.33611E+01
11.33	.29018E+00	.39875E+02	.34855E+01
11.37	.24572E+00	.39958E+C2	.35895E+01
11.41	.20800E+00	.40041E+02	.36762E+01
11.46	.17601E+00	.40125E+02	.37483E+01
11.50	.14888E+00	.40208E+02	.38080E+01
11.54	.12563E+00	.41318E+02	.37620E+01
11.58	.10551E+00	.42425E+02	.37112E+01
11.62	.88216E-01	.43524E+02	.36572E+01
11.66	.73419E-01	.44612E+02	.36012E+01
11.71	.60828E-01	.45685E+02	.35442E+01
11.75	.50174E-01	.46741E+02	.34869E+01
11.79	.41206E-01	.47776E+02	.34301E+01
11.83	.33697E-01	.48790E+02	.33742E+01
11.87	.27441E-01	.49781E+02	.33197E+01
11.91	.22256E-01	.50747E+02	.32667E+01
11.96	.17979E-01	.51688E+02	.32155E+01
12.00	.14468E-01	.52604E+02	.31662E+01
12.04	.11599E-01	.53494E+02	.31189E+01
12.08	.92646E-02	.54358E+02	.30736E+01
12.12	.73740E-02	.55196E+02	.30303E+01
12.16	.58491E-02	.56009E+02	.29891E+01
12.21	.46241E-02	.56797E+02	.29498E+01
12.25	.36438E-02	.57559E+02	.29124E+01
12.29	.28624E-02	.58298E+02	.28769E+01
12.33	.22418E-02	.59012E+02	.28431E+01
12.37	.17506E-02	.59704E+02	.28110E+01
12.41	.13632E-02	.60373E+02	.27805E+01
12.46	.10586E-02	.61020E+02	.27515E+01
12.50	.81984E-03	.61645E+02	.27239E+01
12.54	.63334E-03	.62250E+02	.26978E+01
12.58	.48805E-03	.62834E+02	.26729E+01
12.62	.37519E-03	.63400E+02	.26493E+01
12.66	.28776E-03	.63946E+02	.26268E+01
12.71	.22022E-03	.64474E+02	.26054E+01
12.75	.16816E-03	.64984E+02	.25850E+01
12.79	.12814E-03	.65477E+02	.25656E+01
12.83	.97448E-04	.65954E+02	.25471E+01
12.87	.73963E-04	.66414E+02	.25295E+01
12.91	.56032E-04	.66859E+02	.25127E+01
12.96	.42371E-04	.67289E+02	.24966E+01
13.00	.31984E-04	.67705E+02	.24813E+01
13.04	.24102E-04	.68106E+02	.24667E+01
13.08	.18133E-04	.68495E+02	.24527E+01
13.12	.13620E-04	.68870E+02	.24394E+01
13.16	.10215E-04	.69232E+02	.24266E+01
13.21	.76498E-05	.69582E+02	.24144E+01
13.25	.57205E-05	.69920E+02	.24027E+01
13.29	.42719E-05	.70247E+02	.23915E+01

Table C-3. Continued.

EQUATION SET #3
RESULTS OF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS30

TIME	N	X	PERCENT N
13.33	.31858E-05	.70563E+02	.23808E+01
13.37	.23728E-05	.70869E+02	.23706E+01
13.41	.17651E-05	.71164E+02	.23608E+01
13.46	.13114E-05	.71449E+02	.23513E+01
13.50	.97320E-06	.71724E+02	.23423E+01
13.54	.72141E-06	.71990E+02	.23336E+01
13.58	.53418E-06	.72248E+02	.23253E+01
13.62	.39512E-06	.72496E+02	.23174E+01
13.66	.29197E-06	.72736E+02	.23097E+01
13.71	.21553E-06	.72968E+02	.23024E+01
13.75	.15896E-06	.73192E+02	.22953E+01
13.79	.11712E-06	.73409E+02	.22885E+01
13.83	.86223E-07	.73619E+02	.22820E+01
13.87	.63420E-07	.73821E+02	.22758E+01
13.91	.46610E-07	.74016E+02	.22698E+01
13.96	.34228E-07	.74205E+02	.22640E+01
14.00	.25115E-07	.74388E+02	.22584E+01
14.04	.18415E-07	.74564E+02	.22531E+01
14.08	.13493E-07	.74735E+02	.22480E+01
14.12	.98794E-08	.74899E+02	.22430E+01
14.16	.72287E-08	.75058E+02	.22383E+01
14.21	.52857E-08	.75212E+02	.22337E+01
14.25	.38626E-08	.75361E+02	.22293E+01
14.29	.28209E-08	.75504E+02	.22250E+01
14.33	.20589E-08	.75643E+02	.22210E+01
14.37	.15019E-08	.75777E+02	.22170E+01
14.41	.10950E-08	.75907E+02	.22132E+01
14.46	.79791E-09	.76032E+02	.22096E+01
14.50	.58113E-09	.76153E+02	.22061E+01
14.54	.42303E-09	.76270E+02	.22027E+01
14.58	.30780E-09	.76383E+02	.21994E+01
14.62	.22385E-09	.76492E+02	.21963E+01
14.66	.16272E-09	.76597E+02	.21933E+01
14.71	.11824E-09	.76699E+02	.21904E+01
14.75	.85880E-10	.76798E+02	.21876E+01
14.79	.62351E-10	.76893E+02	.21849E+01
14.83	.45251E-10	.76985E+02	.21822E+01
14.87	.32828E-10	.77074E+02	.21797E+01
14.91	.23807E-10	.77160E+02	.21773E+01
14.96	.17259E-10	.77243E+02	.21750E+01
15.00	.12508E-10	.77323E+02	.21727E+01
15.04	.90616E-11	.77400E+02	.21705E+01
15.08	.65628E-11	.77475E+02	.21684E+01
15.12	.47516E-11	.77547E+02	.21664E+01
15.16	.34392E-11	.77617E+02	.21645E+01
15.21	.24886E-11	.77685E+02	.21626E+01
15.25	.18002E-11	.77750E+02	.21608E+01
15.29	.13019E-11	.77813E+02	.21590E+01
15.33	.94132E-12	.77874E+02	.21573E+01
15.37	.68042E-12	.77933E+02	.21557E+01
15.41	.49171E-12	.77990E+02	.21541E+01
15.46	.35526E-12	.78045E+02	.21526E+01
15.50	.25662E-12	.78098E+02	.21511E+01
15.54	.18532E-12	.78149E+02	.21497E+01

Table C-3. Continued.

EQUATION SET #3
RESULTS CF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS 30

TIME	N	X	PERCENT N
15.58	.13381E-12	.78199E+02	.21484E+01
15.62	.96591E-13	.78247E+02	.21471E+01
15.66	.69713E-13	.78293E+02	.21458E+01
15.71	.56478E+00	.39292E+02	.24566E+01
15.75	.47922E+00	.39580E+02	.26548E+01
15.79	.40629E+00	.39662E+02	.28332E+01
15.83	.34435E+00	.39745E+02	.29832E+01
15.87	.29174E+00	.39828E+02	.31090E+01
15.91	.24709E+00	.39911E+02	.32144E+01
15.96	.20920E+00	.39994E+02	.33025E+01
16.00	.17706E+00	.40077E+02	.33758E+01
16.04	.14980E+00	.40161E+02	.34367E+01
16.08	.12670E+00	.40245E+02	.34869E+01
16.12	.10712E+00	.40329E+02	.35282E+01
16.16	.90535E-01	.40413E+02	.35619E+01
16.21	.76491E-01	.40497E+02	.35892E+01
16.25	.64603E-01	.40582E+02	.36110E+01
16.29	.54544E-01	.40666E+02	.36282E+01
16.33	.46034E-01	.40751E+02	.36415E+01
16.37	.38838E-01	.40836E+02	.36516E+01
16.41	.32756E-01	.40921E+02	.36589E+01
16.46	.27616E-01	.41006E+02	.36638E+01
16.50	.23275E-01	.41092E+02	.36667E+01
16.54	.19609E-01	.41178E+02	.36680E+01
16.58	.16514E-01	.41264E+02	.36679E+01
16.62	.13876E-01	.42293E+02	.35848E+01
16.66	.11610E-01	.43291E+02	.35074E+01
16.71	.96740E-02	.44259E+02	.34351E+01
16.75	.80291E-02	.45197E+02	.33674E+01
16.79	.66383E-02	.46106E+02	.33040E+01
16.83	.54679E-02	.46986E+02	.32446E+01
16.87	.60220E+00	.24134E+02	.38443E+01
16.91	.54379E+00	.24850E+02	.39686E+01
16.96	.48952E+00	.25632E+02	.40593E+01
17.00	.43917E+00	.26471E+02	.41208E+01
17.04	.39259E+00	.27360E+02	.41573E+01
17.08	.34962E+00	.28289E+02	.41725E+01
17.12	.31012E+00	.29254E+02	.41700E+01
17.16	.27397E+00	.30246E+02	.41527E+01
17.21	.24102E+00	.31260E+02	.41234E+01
17.25	.21114E+00	.32289E+02	.40845E+01
17.29	.18416E+00	.33330E+02	.40379E+01
17.33	.15993E+00	.34376E+02	.39855E+01
17.37	.13829E+00	.35424E+02	.39287E+01
17.41	.11905E+00	.36469E+02	.38689E+01
17.46	.10205E+00	.37508E+02	.38071E+01
17.50	.87098E-01	.38537E+02	.37442E+01
17.54	.74021E-01	.39553E+02	.36810E+01
17.58	.62643E-01	.40555E+02	.36181E+01
17.62	.52795E-01	.41541E+02	.35560E+01
17.66	.44315E-01	.42507E+02	.34951E+01
17.71	.37048E-01	.43454E+02	.34357E+01
17.75	.30853E-01	.44379E+02	.33780E+01
17.79	.25596E-01	.45282E+02	.33223E+01

Table C-3. Continued.

EQUATION SET #3
RESULTS CF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS 30

TIME	N	X	PERCENT N
17.83	.21156E-01	.46163E+02	.32685E+01
17.87	.17423E-01	.47020E+02	.32169E+01
17.91	.14298E-01	.47854E+02	.31673E+01
17.96	.11694E-01	.48665E+02	.31199E+01
18.00	.95319E-02	.49452E+02	.30747E+01
18.04	.77446E-02	.50215E+02	.30315E+01
18.08	.62728E-02	.50956E+02	.29903E+01
18.12	.50653E-02	.51673E+02	.29511E+01
18.16	.40782E-02	.52368E+02	.29138E+01
18.21	.32741E-02	.53041E+02	.28784E+01
18.25	.26214E-02	.53693E+02	.28446E+01
18.29	.20931E-02	.54324E+02	.28126E+01
18.33	.16670E-02	.54934E+02	.27821E+01
18.37	.13244E-02	.55524E+02	.27532E+01
18.41	.10496E-02	.56095E+02	.27257E+01
18.46	.82987E-03	.56646E+02	.26995E+01
18.50	.65467E-03	.57180E+02	.26746E+01
18.54	.51533E-03	.57696E+02	.26509E+01
18.58	.40479E-03	.58194E+02	.26284E+01
18.62	.31732E-03	.58677E+02	.26070E+01
18.66	.24825E-03	.59142E+02	.25866E+01
18.71	.19385E-03	.59593E+02	.25671E+01
18.75	.15109E-03	.60028E+02	.25486E+01
18.79	.11755E-03	.60448E+02	.25309E+01
18.83	.91303E-04	.60855E+02	.25140E+01
18.87	.70796E-04	.61248E+02	.24979E+01
18.91	.54807E-04	.61627E+02	.24826E+01
18.96	.42364E-04	.61994E+02	.24679E+01
19.00	.32696E-04	.62349E+02	.24539E+01
19.04	.25198E-04	.62691E+02	.24405E+01
19.08	.19392E-04	.63022E+02	.24277E+01
19.12	.14904E-04	.63342E+02	.24154E+01
19.16	.11439E-04	.63651E+02	.24037E+01
19.21	.87690E-05	.63950E+02	.23925E+01
19.25	.67139E-05	.64238E+02	.23818E+01
19.29	.51343E-05	.64517E+02	.23715E+01

Table C-4. Simulation of dilutions of experiment #2, transition point modified to 3.0 percent. (See
Figures 19 - 21.)

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OESERVEO EXPERMENTAL DATA          MEAN TEMP=24.9      RANGE  22.0 TO 26.0
MEAN VALUES OF RUN 3               MEAN PH 7.1         RANGE   6.9 TO  8.5
                                                      TCTAL NITROGEN= 1.57
                      DRY    CELLULAR   EXTERNAL
            TIME      WT.        N          N
           (DAYS)   (MG/L)    (MG/L)     (MG/L)
            0.93     3.13      0.19       1.38
            1.50     9.77      0.69       0.88
            2.00    23.40      1.26       0.31
            3.05    47.33      1.57       0.00
            4.03    58.20      1.56       0.01
            5.01    69.27      1.57      -0.00
           11.04    79.00      1.68      -0.11
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Table C-4. Continued.

EQUATION SET #3

DN/DT=-G*N*X   DX/DT=U*(PERN-NMIN)*X   DILUTION ROUTINE (OPTIONAL)
N=EXTRACELLULAR NITROGEN (MG/L)   N=D(1)*N+D(2)   X=D(3)*X+D(4)
X=ALGAL BIOMASS AS DRY WT. (MG/L)   C(6)=D(9)
PERN=PERCENT NITROGEN=(NTOT-N)/X
NMIN=MINIMUM PERCENT NITROGEN FOR POSITIVE GROWTH
NTOT=TOTAL NITROGEN (MG/L)   C(1)=G   C(2)=U   C(6)=NTOT   C(7)=NMIN

                              DEFINITION OF CONSTANTS
C(1)= .100E+00   C(2)= .390E+00   C(3)= .500E-01   C(4)= .380E+01   C(5)=0.   C(6)= .157E+01   C(7)= .211E+01
                              DEFINITION OF DILUTION CONSTANTS
                              TIME OF DILUTION= 0.00
D(1)= 0.000   D(2)= 0.000   D(3)= 0.000   D(4)= 0.000   D(5)= 0.000   D(6)= 0.000   D(7)= 0.000   D(8)= 0.000   D(9)= 0.000

Table C-4. Continued.

EQUATION SET #3
RESULTS OF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS 30

TIME	N	X	PERCENT N
0.93	.13841E+01	.31333E+01	.59339E+01
0.99	.13655E+01	.33395E+01	.61224E+01
1.03	.13460E+01	.35691E+01	.62750E+01
1.08	.13255E+01	.38229E+01	.63961E+01
1.12	.13038E+01	.41019E+01	.64899E+01
1.16	.12809E+01	.44072E+01	.65600E+01
1.20	.12567E+01	.47397E+01	.66096E+01
1.24	.12312E+01	.51007E+01	.66415E+01
1.28	.12044E+01	.54913E+01	.66582E+01
1.33	.11761E+01	.59128E+01	.66616E+01
1.37	.11464E+01	.63664E+01	.66534E+01
1.41	.11153E+01	.68532E+01	.66349E+01
1.45	.10827E+01	.73746E+01	.66074E+01
1.49	.10488E+01	.79315E+01	.65717E+01
1.53	.10134E+01	.85250E+01	.65286E+01
1.58	.97680E+00	.91560E+01	.64789E+01
1.62	.93894E+00	.98253E+01	.64229E+01
1.66	.89996E+00	.10533E+02	.63612E+01
1.70	.85999E+00	.11281E+02	.62941E+01
1.74	.81917E+00	.12067E+02	.62220E+01
1.78	.77767E+00	.12893E+02	.61453E+01
1.83	.73568E+00	.13758E+02	.60642E+01
1.87	.69339E+00	.14661E+02	.59791E+01
1.91	.65104E+00	.15601E+02	.58903E+01
1.95	.60883E+00	.16577E+02	.57981E+01
1.99	.56701E+00	.17588E+02	.57028E+01
2.03	.52581E+00	.18630E+02	.56048E+01
2.08	.48545E+00	.19703E+02	.55045E+01
2.12	.44617E+00	.20803E+02	.54022E+01
2.16	.40818E+00	.21928E+02	.52984E+01
2.20	.37165E+00	.23074E+02	.51935E+01
2.24	.33677E+00	.24239E+02	.50878E+01
2.28	.30367E+00	.25419E+02	.49818E+01
2.33	.27248E+00	.26610E+02	.48760E+01
2.37	.24327E+00	.27810E+02	.47707E+01
2.41	.21611E+00	.29014E+02	.46663E+01
2.45	.19103E+00	.30219E+02	.45632E+01
2.49	.16800E+00	.31422E+02	.44618E+01
2.53	.14702E+00	.32620E+02	.43623E+01
2.58	.12802E+00	.33809E+02	.42651E+01
2.62	.11092E+00	.34987E+02	.41703E+01
2.66	.95645E-01	.36151E+02	.40783E+01
2.70	.82074E-01	.37299E+02	.39892E+01
2.74	.70095E-01	.38428E+02	.39031E+01
2.78	.59586E-01	.39538E+02	.38202E+01
2.83	.50421E-01	.40625E+02	.37405E+01
2.87	.42475E-01	.41690E+02	.36640E+01
2.91	.35625E-01	.42731E+02	.35908E+01
2.95	.29751E-01	.43746E+02	.35209E+01
2.99	.24743E-01	.44736E+02	.34541E+01
3.03	.20494E-01	.45700E+02	.33906E+01
3.08	.16907E-01	.46638E+02	.33301E+01
3.12	.13895E-01	.47550E+02	.32726E+01
3.16	.11376E-01	.48435E+02	.32180E+01

Table C-4. Continued.

EQUATION SET #3
RESULTS OF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS 30

TIME	N	X	PERCENT N
3.20	.92807E-02	.49294E+02	.31661E+01
3.24	.75444E-02	.50127E+02	.31170E+01
3.28	.61121E-02	.50935E+02	.30704E+01
3.33	.49353E-02	.51717E+02	.30262E+01
3.37	.39723E-02	.52475E+02	.29843E+01
3.41	.31873E-02	.53208E+02	.29447E+01
3.45	.25497E-02	.53918E+02	.29071E+01
3.49	.20338E-02	.54605E+02	.28715E+01
3.53	.16177E-02	.55270E+02	.28377E+01
3.58	.12832E-02	.55913E+02	.28057E+01
3.62	.10152E-02	.56534E+02	.27753E+01
3.66	.80114E-03	.57135E+02	.27465E+01
3.70	.63066E-03	.57716E+02	.27191E+01
3.74	.49528E-03	.58278E+02	.26931E+01
3.78	.38806E-03	.58821E+02	.26685E+01
3.83	.30338E-03	.59346E+02	.26450E+01
3.87	.23667E-03	.59853E+02	.26227E+01
3.91	.18424E-03	.60343E+02	.26015E+01
3.95	.14314E-03	.60817E+02	.25813E+01
3.99	.11100E-03	.61275E+02	.25620E+01
4.03	.85907E-04	.61717E+02	.25437E+01
4.08	.66369E-04	.62145E+02	.25262E+01
4.12	.51184E-04	.62558E+02	.25096E+01
4.16	.39407E-04	.62958E+02	.24937E+01
4.20	.30290E-04	.63343E+02	.24785E+01
4.24	.23246E-04	.63716E+02	.24640E+01
4.28	.17812E-04	.64077E+02	.24502E+01
4.33	.13629E-04	.64425E+02	.24369E+01
4.37	.10413E-04	.64761E+02	.24243E+01
4.41	.79451E-05	.65086E+02	.24122E+01
4.45	.60540E-05	.65401E+02	.24006E+01
4.49	.46071E-05	.65704E+02	.23895E+01
4.53	.35016E-05	.65998E+02	.23789E+01
4.58	.26582E-05	.66281E+02	.23687E+01
4.62	.20156E-05	.66555E+02	.23589E+01
4.66	.15266E-05	.66820E+02	.23496E+01
4.70	.11550E-05	.67075E+02	.23406E+01
4.74	.87298E-06	.67323E+02	.23321E+01
4.78	.65912E-06	.67561E+02	.23238E+01
4.83	.49717E-06	.67792E+02	.23159E+01
4.87	.37466E-06	.68015E+02	.23083E+01
4.91	.28208E-06	.68231E+02	.23010E+01
4.95	.21219E-06	.68439E+02	.22940E+01
4.99	.15948E-06	.68640E+02	.22873E+01
5.03	.11976E-06	.68834E+02	.22808E+01
5.08	.89866E-07	.69022E+02	.22746E+01
5.12	.67381E-07	.69204E+02	.22687E+01
5.16	.50484E-07	.69379E+02	.22629E+01
5.20	.37798E-07	.69549E+02	.22574E+01
5.24	.28279E-07	.69712E+02	.22521E+01
5.28	.21144E-07	.69871E+02	.22470E+01
5.33	.15799E-07	.70024E+02	.22421E+01
5.37	.11797E-07	.70171E+02	.22374E+01
5.41	.88039E-08	.70314E+02	.22328E+01

Table C-4. Continued.

EQUATION SET #3
RESULTS OF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS 30

TIME	N	X	PERCENT N
5.45	.65663E-08	.70452E+02	.22285E+01
5.49	.48946E-08	.70585E+02	.22243E+01
5.53	.36465E-08	.70714E+02	.22202E+01
5.58	.27153E-08	.70839E+02	.22163E+01
5.62	.20208E-08	.70959E+02	.22125E+01
5.66	.15032E-08	.71075E+02	.22089E+01
5.70	.11177E-08	.71188E+02	.22054E+01
5.74	.83062E-09	.71296E+02	.22021E+01
5.78	.61702E-09	.71401E+02	.21988E+01
5.83	.45815E-09	.71502E+02	.21957E+01
5.87	.34005E-09	.71600E+02	.21927E+01
5.91	.25229E-09	.71695E+02	.21898E+01
5.95	.18711E-09	.71786E+02	.21870E+01
5.99	.13871E-09	.71875E+02	.21844E+01
6.03	.10280E-09	.71960E+02	.21818E+01
6.08	.76155E-10	.72043E+02	.21793E+01
6.12	.56399E-10	.72122E+02	.21769E+01
6.16	.41754E-10	.72199E+02	.21745E+01
6.20	.30902E-10	.72274E+02	.21723E+01
6.24	.22864E-10	.72346E+02	.21701E+01
6.28	.16911E-10	.72415E+02	.21681E+01
6.33	.12505E-10	.72482E+02	.21660E+01
6.37	.92443E-11	.72547E+02	.21641E+01
6.41	.68320E-11	.72610E+02	.21622E+01
6.45	.50478E-11	.72671E+02	.21604E+01
6.49	.37287E-11	.72729E+02	.21587E+01
6.53	.27536E-11	.72786E+02	.21570E+01
6.58	.20331E-11	.72840E+02	.21554E+01
6.62	.15007E-11	.72893E+02	.21538E+01
6.66	.11076E-11	.72944E+02	.21523E+01
6.70	.81721E-12	.72994E+02	.21509E+01
6.74	.60285E-12	.73041E+02	.21495E+01
6.78	.44464E-12	.73087E+02	.21481E+01
6.83	.32788E-12	.73132E+02	.21468E+01
6.87	.24174E-12	.73175E+02	.21455E+01
6.91	.17820E-12	.73216E+02	.21443E+01
6.95	.13134E-12	.73256E+02	.21432E+01
6.99	.96783E-13	.73295E+02	.21420E+01
7.03	.71309E-13	.73333E+02	.21409E+01
7.08	.52531E-13	.73369E+02	.21399E+01
7.12	.38693E-13	.73404E+02	.21388E+01
7.16	.28496E-13	.73438E+02	.21379E+01
7.20	.20983E-13	.73470E+02	.21369E+01
7.24	.15449E-13	.73502E+02	.21360E+01
7.28	.11373E-13	.73533E+02	.21351E+01
7.33	.83713E-14	.73562E+02	.21343E+01
7.37	.61611E-14	.73591E+02	.21334E+01
7.41	.45339E-14	.73618E+02	.21326E+01
7.45	.33361E-14	.73645E+02	.21319E+01
7.49	.24545E-14	.73670E+02	.21311E+01
7.53	.18057E-14	.73695E+02	.21304E+01
7.58	.13282E-14	.73719E+02	.21297E+01
7.62	.97691E-15	.73743E+02	.21290E+01
7.66	.71846E-15	.73765E+02	.21284E+01

Table C-4. Continued.

EQUATION SET #3
RESULTS OF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS30

TIME	N	X	PERCENT N
7.70	.52834E-15	.73787E+02	.21278E+01
7.74	.38849E-15	.73808E+02	.21272E+01
7.78	.28563E-15	.73828E+02	.21266E+01
7.83	.20999E-15	.73847E+02	.21260E+01
7.87	.15437E-15	.73866E+02	.21255E+01
7.91	.11347E-15	.73884E+02	.21249E+01
7.95	.83404E-16	.73902E+02	.21244E+01
7.99	.61298E-16	.73919E+02	.21239E+01
8.03	.45049E-16	.73936E+02	.21235E+01
8.08	.33104E-16	.73951E+02	.21230E+01
8.12	.24325E-16	.73967E+02	.21226E+01
8.16	.17873E-16	.73982E+02	.21221E+01
8.20	.13132E-16	.73996E+02	.21217E+01
8.24	.96477E-17	.74010E+02	.21213E+01
8.28	.70875E-17	.74023E+02	.21210E+01
8.33	.52065E-17	.74036E+02	.21206E+01
8.37	.38244E-17	.74049E+02	.21202E+01
8.41	.28091E-17	.74061E+02	.21199E+01
8.45	.20632E-17	.74073E+02	.21195E+01
8.49	.15153E-17	.74084E+02	.21192E+01
8.53	.11129E-17	.74095E+02	.21189E+01
8.58	.81728E-18	.74105E+02	.21186E+01
8.62	.60016E-18	.74116E+02	.21183E+01
8.66	.44071E-18	.74125E+02	.21180E+01
8.70	.32361E-18	.74135E+02	.21178E+01
8.74	.23761E-18	.74144E+02	.21175E+01
8.78	.17446E-18	.74153E+02	.21172E+01
8.83	.12809E-18	.74162E+02	.21170E+01
8.87	.94041E-19	.74170E+02	.21168E+01
8.91	.69041E-19	.74178E+02	.21165E+01
8.95	.50685E-19	.74186E+02	.21163E+01
8.99	.37208E-19	.74193E+02	.21161E+01
9.03	.27314E-19	.74200E+02	.21159E+01
9.08	.20050E-19	.74207E+02	.21157E+01
9.12	.14718E-19	.74214E+02	.21155E+01
9.16	.10803E-19	.74221E+02	.21153E+01
9.20	.79294E-20	.74227E+02	.21151E+01
9.24	.58200E-20	.74233E+02	.21150E+01
9.28	.42717E-20	.74239E+02	.21148E+01
9.33	.31352E-20	.74245E+02	.21146E+01
9.37	.23010E-20	.74250E+02	.21145E+01
9.41	.16887E-20	.74255E+02	.21143E+01
9.45	.12394E-20	.74260E+02	.21142E+01
9.49	.90954E-21	.74265E+02	.21140E+01
9.53	.66748E-21	.74270E+02	.21139E+01
9.58	.48983E-21	.74275E+02	.21138E+01
9.62	.35946E-21	.74279E+02	.21136E+01
9.66	.26378E-21	.74284E+02	.21135E+01
9.70	.19356E-21	.74288E+02	.21134E+01
9.74	.14204E-21	.74292E+02	.21133E+01
9.78	.10422E-21	.74296E+02	.21132E+01
9.83	.76477E-22	.74300E+02	.21131E+01
9.87	.56116E-22	.74303E+02	.21130E+01
9.91	.41175E-22	.74307E+02	.21129E+01

Table C-4.   Continued.

EQUATION SET #3
RESULTS OF ALGAL GROWTH SIMULATION
NUMBER OF MINUTES IN A TIME STEP IS 30

TIME	N	X	PERCENT N
9.95	.30212E-22	.74310E+02	.21128E+01
9.99	.22168E-22	.74313E+02	.21127E+01
10.03	.16265E-22	.74317E+02	.21126E+01
10.08	.11934E-22	.74320E+02	.21125E+01
10.12	.87558E-23	.74323E+02	.21124E+01
10.16	.64241E-23	.74325E+02	.21123E+01
10.20	.47132E-23	.74328E+02	.21123E+01
10.24	.34580E-23	.74331E+02	.21122E+01
10.28	.25370E-23	.74333E+02	.21121E+01
10.33	.18613E-23	.74336E+02	.21120E+01
10.37	.13656E-23	.74338E+02	.21120E+01
10.41	.10018E-23	.74341E+02	.21119E+01
10.45	.73499E-24	.74343E+02	.21118E+01
10.49	.53921E-24	.74345E+02	.21118E+01
10.53	.39558E-24	.74347E+02	.21117E+01
10.58	.29021E-24	.74349E+02	.21117E+01
10.62	.21290E-24	.74351E+02	.21116E+01
10.66	.15618E-24	.74353E+02	.21115E+01
10.70	.11458E-24	.74355E+02	.21115E+01
10.74	.84054E-25	.74357E+02	.21114E+01
10.78	.61661E-25	.74358E+02	.21114E+01
10.83	.45234E-25	.74360E+02	.21113E+01
10.87	.33183E-25	.74362E+02	.21113E+01
10.91	.24342E-25	.74363E+02	.21113E+01
10.95	.17857E-25	.74365E+02	.21112E+01
10.99	.13099E-25	.74366E+02	.21112E+01
11.03	.96089E-26	.74368E+02	.21111E+01
11.08	.70487E-26	.74369E+02	.21111E+01

VITA

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