

A STUDY OF NUTRIENT ACCRUAL, UPTAKE,
AND REGENERATION AS RELATED TO
PRIMARY PRODUCTION IN A
WARM-WATER STREAM

Thesis for the Degree of Ph. D.
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Morris Leroy Brehmer
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AS RELATED TO PRIMARY PRODUCTION IN A WARM-WATER STREAM

presented by

MORRIS LEROY BREHMER

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by

MORRIS LEROY BREHMER

AN ABSTRACT

Submitted to the School for Advanced Graduate Studies of
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Robert C. Ball

The Red Cedar River, a warm-water stream which drains a portion of the south-central part of the lower peninsula of Michigan, was investigated to determine the major nutrient sources and the relationship between primary production and nutrient levels.

The results of a study of the entire stream indicated that the major changes in nutrient levels occurred after the introduction of effluents from municipal drains and sewage treatment plant outfalls. The nutrient contributions from the tributary streams were of the greatest magnitude during periods of high run-off from the watershed area and had little effect on the nutrient budget of the main stream.

An intensive study was made on a 5.3 mile area of the river to determine the rate of nutrient accrual, uptake, and regeneration as related to the seasonal primary production patterns. Upstream from this area a reservoir served as a silt basin, just within the upper limits a turbid tributary stream emptied into the river, and within the first 0.5 mile the outfall from a sewage treatment plant provided a continuous source of nutrients without producing septic conditions.

The dissolved, sestonic, acid-soluble sestonic, and total phosphorus, the ammonia and nitrite plus nitrate nitrogen, and the periphyton production were determined at nine stations within the 5.3 mile area during a 13-month period. The periphyton measurements involved the use of plexiglass artificial substrata. The optical absorbancy of the ethanol-extracted phytopigments from the periphyton accumulation on the substrata was used as an index of production.

The data indicated that during average water-level conditions the stream was enriched by more than $100 \mu\text{g l}^{-1}$ of phosphorus and 0.5 mg l^{-1} of inorganic nitrogen by the effluent from the sewage treatment plant. The flora of the stream removed nearly all of the added nutrients from solution within the first 0.6 mile downstream from the outfall during all periods of the year except when ice cover was present. This occurred even during the summer months when the periphyton growth appeared to be inhibited by unidentified agents introduced into the stream with the effluent. The organic phosphorus:phytopigment density ratio in the periphyton was found to be more than four times greater at the station 0.3 mile downstream from the outfall than in the areas upstream from the outfall.

During the summer months the dissolved phosphorus content of the water decreased rapidly within the first 0.6 mile downstream from the outfall and then increased toward the downstream stations. The area of phosphorus regeneration was dependent upon stream flow and water temperature.

The data indicate that periphyton production increased rapidly after the spring thaw and reached a maximum in April or early May. The production then decreased gradually until an extraneous nitrogen supply produced a second maximum in June. After the June peak the production decreased gradually until the period of ice cover. The production levels appear to be more limited by the inorganic nitrogen content of the water than by the dissolved phosphorus content.

The standing crop of periphyton in the stream was almost completely destroyed by high water and associated high turbidities which existed for only a short period of time.

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INTRODUCTION

INTRODUCTION

One of the most difficult problems facing the aquatic ecologist is the finding of a method for determining and measuring quantitatively the physiochemical and biological processes operating in a lotic environment. A stream is a more open ecosystem than a lake and the factors controlling the biological activity may be more closely related to the watershed than to the stream bed. Also, the unidirectional flow continually carries the seston away from the area in which it was produced so that the organically combined nutrients are lost from the biotope and the development of an autochthonic plankton population is virtually impossible.

The wide variations in velocity, turbidity, and water level in a stream tend to alter the substratum. The existing flora and fauna can be destroyed by the exclusion of light or by the abrasive action of the bed load associated with high-water conditions. Therefore, it is possible for the entire biological system of a stream to be disrupted by the run-off from a single rain. One can surely agree with Purdy (1923) who said "A large lake represents stability of environment, but a flowing stream is the fullest expression of a condition of instability."

In view of the increasing demands for aquatic recreational areas, the need for a better understanding of stream metabolism and stream production becomes more apparent. Paradoxically, at a time when the

streams are destined to carry a heavier recreational load, they are also receiving more wastes from human activities. Nearly all streams, with the exception of those remote from human habitation, serve to a greater or lesser extent as a dumping ground for biological and industrial wastes produced by man. It is well established that streams have a capacity for self-purification and that the physical, chemical, and biological forces involved in the stabilization of wastes are interrelated and mutually dependent.

Organic material added to a stream undergoes carbonization, nitrification, dephosphorulation, etc., and releases the basic nutrient constituents to solution. If the amount of organic material added to a stream exceeds its capacity for self-purification and stabilization of the putrescible fraction, the dissolved oxygen supply in the water is depleted. This results in the accumulation of unstabilized waste material on the bottom of the stream and the formation of toxic anaerobic decomposition products. The composition of the flora and fauna is then limited to those species that can tolerate highly adverse environmental conditions.

The modern sewage treatment plant concentrates the physical, chemical, and biological forces involved in the stabilization of organic matter into a system of settling basins, aeration tanks or filters, and digestors, and therefore prevents the formation of a septic zone in the body of water receiving the effluent.

The nutrient content of the effluent varies with the type of treatment and efficiency of the sewage treatment plant. Also, the rate of enrichment of the receiving waters varies with the dilution

factor as influenced by the volume of effluent and with the precipitation and run-off in the watershed area of the stream. The rate of biological response to the introduced nutrients is also dependent upon a myriad of adverse physical conditions which are inherent to flowing waters.

The dynamics of the biological phase in enriched streams can be studied by measuring the production at any one or several of the trophic levels. The presence of a delayed increase in production may indicate that toxic or inhibitory agents are present in the effluent which prevent the utilization of the introduced nutrients by the primary producers. The organisms composing any trophic level can be used to study the biological effects of the introduced nutrients and associated organic and inorganic substances found in sewage treatment plant effluent since they are either directly or indirectly dependent upon the primary producers for food.

The difficulties involved with production measurements at a specific level increase with the position of the organisms in the food chain. Generally speaking, the organisms of the higher levels are characterized by changes in food habits during their life history whereas the members of the lower levels usually can be classified as primary producers or herbivores throughout their entire life history.

Fish are mobile and individuals of many species tend to migrate from areas providing protection and cover to areas of food production or food concentration. Also, some primary consumers such as the white sucker (Catostomus commersonnii) which are independent of many of the "side food chains" so characteristic of the carnivores have a wide

range of movement associated with specific phases of their life history. It is also very difficult to obtain a quantitative sample of a fish population. For these reasons the fish population does not lend itself well to a study of production in streams.

The standing crop and growth rate of the benthic fauna population are often used in productivity studies in a lentic environment. Although there may be significant differences in the species composition and number of organisms found in different bottom types or between the littoral or profundal zones of a lake, the number of microhabitats in a stream bottom increases the sampling problems. On the basis of a single uniform riffle, Needham and Usinger (1956) found that 194 samples would be required to give significant figures on total wet weights at the 95 percent confidence level and that total numbers would require 73 samples.

Since productivity involves the unit of time, the measurements are complicated by variations in water level between sampling periods. The detrimental effects of high water on the benthic fauna are well recognized. Allen (1951) reports that a flood on the Horokiwi Stream destroyed 85 percent of the number and 88 percent of the weight of the benthic fauna.

The length of time period required to accurately detect differences in growth of certain organisms can subject the method to the hazard of high water. Trama (personal communication) reports that 33 days were required for Stenonema pulchellum to grow one millimeter in length when reared in the laboratory under optimum conditions.

The phytoplankton population is frequently used in limnological and oceanographical work to study the productivity of standing waters.

Plankton can usually be found in streams; but, as Ruttner (1952) points out, it is impossible to distinguish between eupotamoplankton and tychoplankton doomed to death in the lotic environment. Also, a sudden rain will often flush the plankton from a stream, or conversely, if the stream channel is characterized by swamps and oxbows, may cause a sudden increase in plankton. Butcher (1932) found that a large portion of the plankton present in streams was detached sessile algae in the process of decomposition. For these reasons the phytoplankton population of a stream is not necessarily indicative of stream conditions.

The periphyton (=Aufwuchs) consists of the community of organisms which grows on the stream bed and on submerged objects in the water. Although benthic fauna are frequently found in the mat, they are not considered as a part of this community.

The periphyton plays an important role in the lotic environment because it is virtually the only primary producer in the ecosystem. Also, because of its perpetuity and rapid turnover period, the volume of this material produced annually in a given area is enormous. Even though the organisms of this group are subjected to the same adverse conditions as the benthic fauna or the fish, they are characterized by a very rapid recovery. The production within this community may also be used to study the nutrient levels of the water mass flowing by since these organisms are not equipped with a means of procuring the essential elements from the stream bed. Considering that the primary consumers in a stream are almost entirely dependent upon this community, the production in the higher trophic levels can be estimated by relating the production of the autotrophic organisms in this group (Lindeman, 1942).

Although Hentschel (1916) was apparently the first to employ artificial substrata to study the accumulation of sessile organisms, the method has been virtually overlooked in this country. Butcher (1932, 1947) mounted glass slides in frames and submerged them in English streams to collect sessile algae for both qualitative and quantitative studies. More recently Patrick (1954) devised the well-known "diatometer" for holding glass slides for the collection of diatoms for evaluating stream conditions. A comprehensive review of the literature concerning the use of artificial substrata for the collection of all types of aquatic and terrestrial microorganisms is given by Cooke (1956).

Hooper, Ball, and Hayne (ms) were the first to combine the phytopigment extract method as used by Kreps and Verbinskaya (1930), Harvey (1934), Manning and Juday (1941), and others with the artificial substrata method for estimating periphyton production. This method has been used and refined by several of their students in studies of fundamental productivity in streams, the data of which are given in the Master's theses of Grzenda (1955) and Alexander (1956).

In this method artificial substrata are exposed in a stream for a given period of time, removed, and the phytopigment from the periphyton growth extracted with 95 percent ethanol. The absorbancy of this solution is then determined with a photoelectric colorimeter. The primary production for the period can then be expressed in terms of net phytopigment density per unit area of substratum to compare areas within a given stream or to compare exposure periods, or converted to units of weight (Grzenda¹) by use of an experimentally determined value

¹ Doctorate thesis in preparation. Method reported at the meeting of the American Institute of Biological Sciences, Bloomington, Indiana, 1958.

representing the relationship between phytopigment density and weight of organic material.

After a preliminary study of the entire water course of the Red Cedar River to determine the physical and chemical characteristics of the water, a 5.3 mile area located immediately downstream from the City of Williamston was chosen for extensive study. Upstream from this area a reservoir served as a silt basin, just within the upper limits a turbid tributary stream emptied into the river, and within the first 0.5 mile the outfall from a sewage treatment plant provided a continuous source of nutrients without producing septic conditions.

The intensive study in the relatively short area was made to determine the fate of nutrients introduced into a natural stream and to measure the biological response as indicated by periphyton production to different nutrient levels. During the course of the study the seasonal changes in periphyton production as well as the effects of adverse physical conditions such as high-water levels and associated high turbidities were determined. It is the aim of this study to obtain a better understanding of nutrient metabolism and the production of organic material in a lotic environment.

METHODS AND TECHNICS

METHODS AND TECHNICS

The year-around sampling program required that the water samples for chemical analyses be cooled in summer and prevented from freezing in winter to preserve the chemical and biological equilibria. This was accomplished by placing the samples in ice water in a portable ice chest immediately after collection in polyethylene bottles. The analyses were either completed the same day or worked to a point where storage would not result in either a gain or loss of constituents.

All dimensional units are given according to the algebraic exponential system. For example, $\frac{1}{\text{g}^2} = \text{g}^{-2}$ and $\text{g}/\text{dm}^3 = \text{g dm}^{-3}$.

Water Temperature

Water temperatures reported for individual stations were taken with a pocket thermometer held approximately three inches under the water surface. The temperatures reported in Appendix A were recorded on a Taylor recording thermometer permanently located ten miles upstream from the mouth of the river and ten miles downstream from Williamston.

pH

The pH values were determined on a Beckman Model N portable pH meter. All measurements were made in the field during the summer period or immediately after returning to the laboratory during the winter months.

Conductivity

The electrical resistance of the water was determined with an Industrial Instrument Company Model RC-7 portable conductivity meter. The resistance readings were corrected to 18° C. and converted to ohms⁻¹ cm⁻¹ x 10⁻⁴. All measurements were made at the sampling location during the summer period and immediately after returning to the laboratory during the winter.

Turbidity

The turbidity measurements were made immediately after returning to the laboratory on a Klett-Summerson photoelectric colorimeter which had been calibrated with the Jackson Candle Turbidimeter. A correction for the intrinsic color of the water was made by adjusting the instrument to zero with a filtered river water sample in the light path. All readings were taken using the blue filter having an approximate spectral range of 400 to 465 millimicrons.

Alkalinity

The alkalinity determinations were made in the laboratory using methods described in "Standard Methods for the Examination of Water, Sewage, and Industrial Wastes" (APHA, AWWA, FSIWA, 1955).

Carbon Dioxide

The free carbon dioxide was determined from the pH and alkalinity readings using the nomograph proposed by Moore (1939).

Dissolved Oxygen

The dissolved oxygen was measured by the unmodified Winkler method. The reagents were added in the field but the final titration was carried out in the laboratory. With few exceptions the samples were taken between 8 a.m. and 11 a.m. Thus the values represent a period when the levels are consistently low due to plant respiration.

Phosphorus

Four physical states of phosphorus were determined during the course of this investigation. In all instances the samples were digested and the phosphorus converted to the PO_4 form, treated with acidified ammonium molybdate, and the density of the blue color resulting from the reduction of the phosphomolybdate with stannous chloride read on a Klett-Summerson colorimeter. The method was modified slightly from that described in Ellis, Westfall, and Ellis (1948) in that the final 100 ml solution was divided and neutralized with saturated NaOH before the final color-producing reagents were added (Taylor, 1937).

Total Phosphorus

The total phosphorus values were obtained after the digestion and treatment of a 100 ml sample of river water.

Total Dissolved Phosphorus

The total dissolved phosphorus was determined from a 100 ml water sample that had been filtered through a Millipore Filter. The HA type membrane having a pore size of 0.45 micron was used for all filtrations.

Sestonic Phosphorus

The sestonic phosphorus values were obtained by difference between the total phosphorus and the dissolved phosphorus.

Acid-Soluble Sestonic Phosphorus

The acid-soluble sestonic phosphorus values were obtained after the digestion and treatment of a 100 ml sample of 0.01 N H_2SO_4 which had been filtered through the pad which retained the seston from the dissolved phosphorus determination.

Early in the program, while becoming familiar with the methods, it was found that the values obtained from samples that had been stored for several days were significantly lower than those obtained from duplicate samples that were analyzed immediately after collection. An attempt was made to determine the mechanism by which the phosphorus was lost from solution and to establish methods of prevention of the loss during storage.

Five liters of water were collected from the Red Cedar River at Williamston, Michigan in a "pyrex" Florence flask. The sample was brought into the laboratory and mixed for three hours on a "Mag-Mix" stirrer to allow for temperature adjustment and to insure the withdrawal of representative sub-samples. The characteristics of the water were as follows:

pH	7.80
Methyl orange alkalinity	256 mg l ⁻¹
Carbon dioxide content	7.8 mg l ⁻¹
Conductivity	612 x 10 ⁻⁴ ohms ⁻¹ cm ⁻¹
Turbidity	17 units.

Ten 100 ml samples were siphoned off and transferred to Erlenmeyer flasks for immediate phosphorus determinations. Ten 4 oz. polyethylene bottles were filled for storage without acid, and ten 4 oz. polyethylene bottles were filled for storage after the addition of 0.3 ml of concentrated sulfuric acid. The samples were stored in the dark at room temperature. The total phosphorus determinations were made according to the stannous chloride-molybdate method previously described.

The results obtained from the phosphorus determinations on the unstored water sample and those stored for 30 days with and without the addition of the acid are given in Table 1.

TABLE 1. Phosphorus values ($\mu\text{g l}^{-1}$) obtained initially and after 30 days storage with and without acidification.

<u>Sample No.</u>	<u>"0" Days</u>	<u>30 Days, Acid Added</u>	<u>30 Days, No Acid</u>
1	55	55	45
2	54	62	22
3	55	56	20
4	54	65	28
5	53	54	39
6	58	55	31
7	54	58	40
8	54	58	32
9	57	54	33
<u>10</u>	<u>54</u>	<u>53</u>	<u>26</u>
Mean Value -	54.8 $\mu\text{g l}^{-1}$	57.0 $\mu\text{g l}^{-1}$	31.6 $\mu\text{g l}^{-1}$

The results of the determinations indicate a significant loss of phosphorus from solution in the untreated samples. In order to determine

if the phosphorus remaining in the storage bottles could be returned to solution or "stripped" from the walls, 100 ml of distilled water and 0.3 ml of concentrated sulfuric acid were added to each of the "no-acid" storage bottles and allowed to stand for 24 hours. Phosphorus determinations on the acidified solutions yielded 22, 12, 23, 18, 28, 20, 17, 16, and 18 $\mu\text{g l}^{-1}$. One sample was lost during handling. The mean recovery value was 19.3 $\mu\text{g l}^{-1}$.

The mean recovery value, when added to the mean value for the samples stored without acid, indicates that phosphorus escaping from solution can be recovered with the addition of acid.

In order to determine if the loss of phosphorus from solution might be due to adsorption on the walls of the bottles under alkaline conditions, two 50 ml samples of a standard phosphate solution were made basic (pH 11.8) by the addition of one drop of NaOH and stored. Determinations made 28 days later indicated that no phosphorus was lost from solution.

At a later date the procedure was repeated and an additional set of samples was stored after 1.0 ml of reagent grade chloroform was added to each bottle according to methods described by Dobie and Moyle (1956). Total phosphorus determinations made 15 days later indicated an average of 12 percent of the phosphorus was lost from solution during the storage while the values obtained from those preserved with 0.3 ml of concentrated sulfuric acid agreed with the original values to within the precision of the method.

Later, Hepher (1958) used phosphorus fortified tap water and found no chemical changes occurred in a sealed jar on storage, but in an

unsealed jar the pH increased, bicarbonate alkalinity decreased, and the carbonate alkalinity increased as carbon dioxide was lost from solution. He also found a loss of phosphorus from solution that roughly corresponded to the calculated solubilities of phosphate as related to pH and calcium ion concentration. Therefore, the 0.3 ml of concentrated sulfuric acid added to the sample bottles increases the solubility of the phosphorus by lowering the pH and preventing the formation of carbonate ions.

Ammonia Nitrogen

The nitrogen present in the form of ammonia was determined by the distillation method as described in "Standard Methods" (APHA, AWWA, FSIWA, 1955).

Nitrogen as Nitrite plus Nitrate

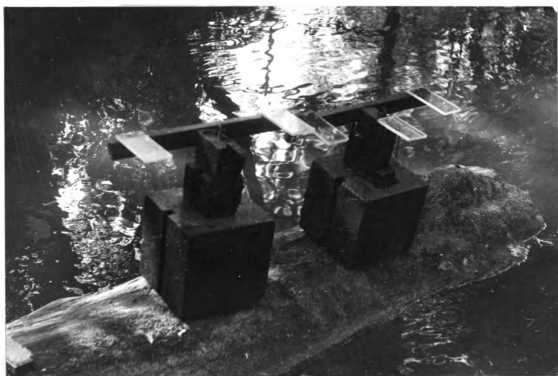
The nitrogen in the oxidized forms was determined by the reduction method as described in "Standard Methods" (APHA, AWWA, FSIWA, 1955).

Periphyton Measurements

The biological sampling program consisted of measuring the periphyton pigments that accumulated on artificial substrata suspended in the stream over a given exposure period. The length of time the substrata were exposed was dependent upon the rate of accumulation as governed by the physical and chemical conditions of the stream.

The substrata employed in this investigation consisted of plexiglass plates, 7 mm in thickness, having an exposed area of 1.4 dm² when attached to a horizontal crossbar (Fig. 1). The basic adaptation

Fig. 1. Plexiglass artificial substrata and supporting blocks
used for the collection of periphyton from the
Red Cedar River.



and method of attachment was devised with a fellow graduate student, Mr. Alfred R. Grzenda.

The plexiglass substrata were collected after the periphyton growth was plainly visible but before a dense mat had formed that would be subject to sloughing due to a layer of dead cells adjacent to the plastic or to the formation of gas bubbles under the mat. The substrata were removed from the stream, placed in individual plastic bags, and frozen to aid in the release of the biological growth from the plastic and to rupture the plant cells to facilitate the phytopigment extraction. The periphyton growth was scraped from the substrata and allowed to stand in 95 percent ethanol for a minimum of 48 hours while stored in total darkness. Tests indicate that samples can be stored in this manner for as long as 30 days without a loss of phytopigments due to decomposition. The samples were filtered through glass wool and the volume of filtrate adjusted to 50 ml by either dilution or evaporation. The color density of the ethanol-soluble phytopigment solution was read on a Klett-Summerson colorimeter using the red filter (640-700 m μ).

Experiments dealing with the opticochemical characteristics of 95 percent ethanol phytopigment extracts show the absorbancy of broad spectrum light (640-700 m μ) is not lineally related to the concentration of the pigments except at very low values. The deviation becomes apparent at approximately 100 units when read on a Klett-Summerson colorimeter and increases proportionately with higher concentrations. This deviation from the Lambert-Beer Law may be due to an interaction between the solvent and the solute or to changes within or among the molecules. This flattening of the curve when absorbancy is plotted against phytopigment concentration destroys the correlation between measured pigment

density and the weight of the organic material from which the pigments were extracted (Grzenda, unpublished).

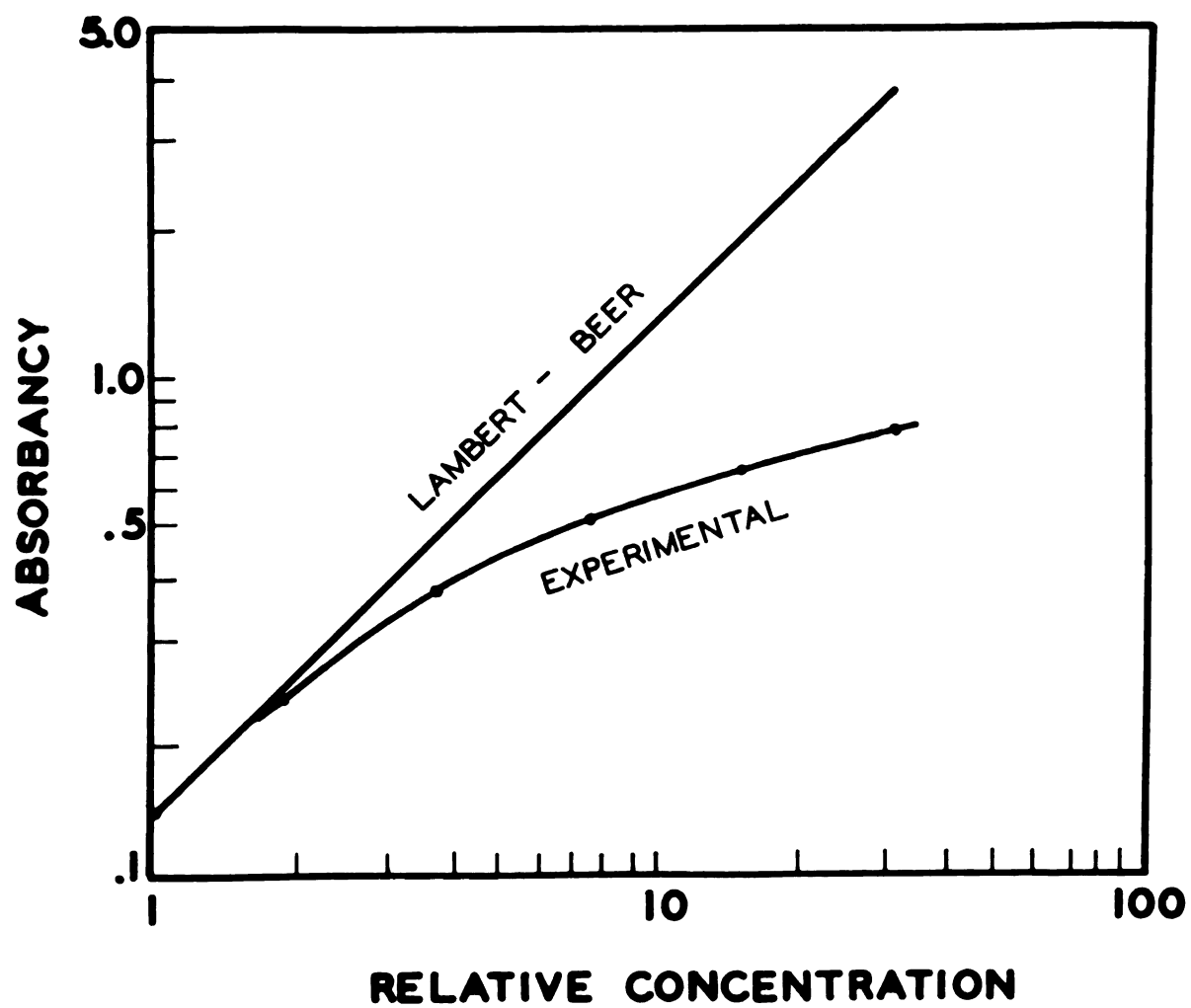
The measured pigment density may be corrected to correspond with the theoretical absorbancy as related to concentration, and the correlation with the weight of organic material restored by determining the deviation from the Lambert-Beer Law. A correction graph was constructed by plotting absorbancy versus concentration as determined by dilution and by concentration of a phyt pigment solution (Fig. 2). In making the correction the measured density is found on the ordinate, followed across to the intercept with the experimentally determined line, and read vertically to the intercept with the extrapolated straight line. The absorbancy unit opposite this intercept represents the corrected reading.

In order to avoid confusion between the measured and the corrected absorbancy values, the unit of adjusted absorbancy, designated as (AA), was adopted for the latter values. The values were then multiplied by 10^3 to avoid the use of the decimal point. Therefore, the corrected absorbancy units of the extracted phyt pigments are given as $AA \times 10^3$. Since these units are lineally related to the weight of periphyton produced, the term phyt pigment unit (optical density or absorbancy of one $AA \times 10^3$) is used as the index of organic material production.

Spectrophotometric Analyses of the Phyt pigments

Periodically, qualitative spectrophotometric analyses were made of the phyt pigment extracts from stations located above and below the sewage treatment outfall. The pigments from a portion of the periphyton

**Fig. 2. Correction graph for adjusting measured phytopigment
absorbancy values to units related to concentration.**



from a substratum were extracted with 90 percent redistilled acetone (Richards with Thompson, 1952) and the absorbancy spectrum plotted from 400 to 700 millimicrons.

DESCRIPTION OF THE AREA

DESCRIPTION OF THE AREA

Physiography

The Red Cedar River, a warm-water stream, is a tributary of the Grand River system and drains the south-central portion of the Lower Peninsula of Michigan. The stream originates as the outflow from Cedar Lake located in Marion Township, Livingston County in Sections 28 and 29, Township 1 North, Range 3 East of the Michigan Meridian and flows through or near the communities of Fowlerville, Webberville, Williamston, and Okemos before entering the East Lansing and Lansing areas and the confluence with the Grand River (Fig. 3). The total length of the main channel is approximately 49 miles and the drainage area is approximately 475 square miles. The river has a mean gradient of 2.5 feet per mile with one-half of the fall occurring in the upper third of the channel.

There are three dams which form artificial impoundments on the Red Cedar River. The largest, located within the City Limits of Williamston, was originally constructed in 1840 to provide water power for a sawmill. The present structure creates a 13 foot working-head of water to operate a constant-flow generator to provide electrical power for a private refrigeration and frozen food plant. The pool created by the Williamston Dam is approximately two miles in length, but for the most part is contained within a narrow belt along the main channel.

Fig. 3. Map of Red Cedar River and principal tributaries. Overlay shows location of the 12 sampling stations used in 1956 to determine the physical and chemical characteristics of the river.

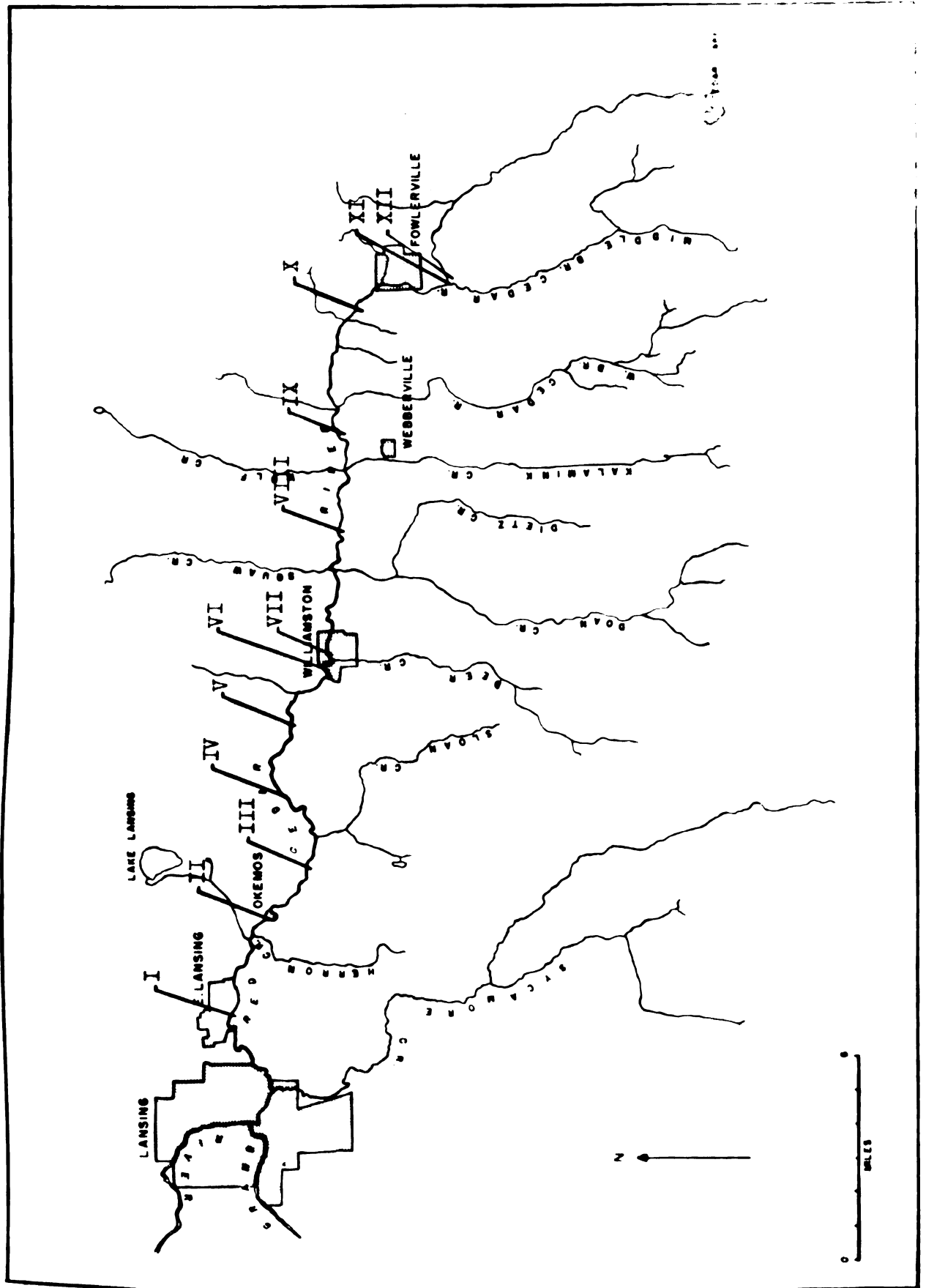
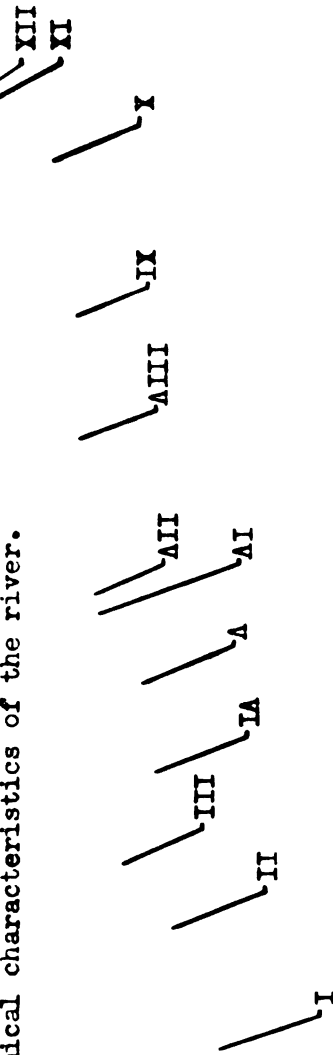


Figure 1. - Red Cedar River and Principal Tributaries

Fig. 3. Map of Red Cedar River and principal tributaries. Overlay shows location of the 12 sampling stations used in 1956 to determine the physical and chemical characteristics of the river.



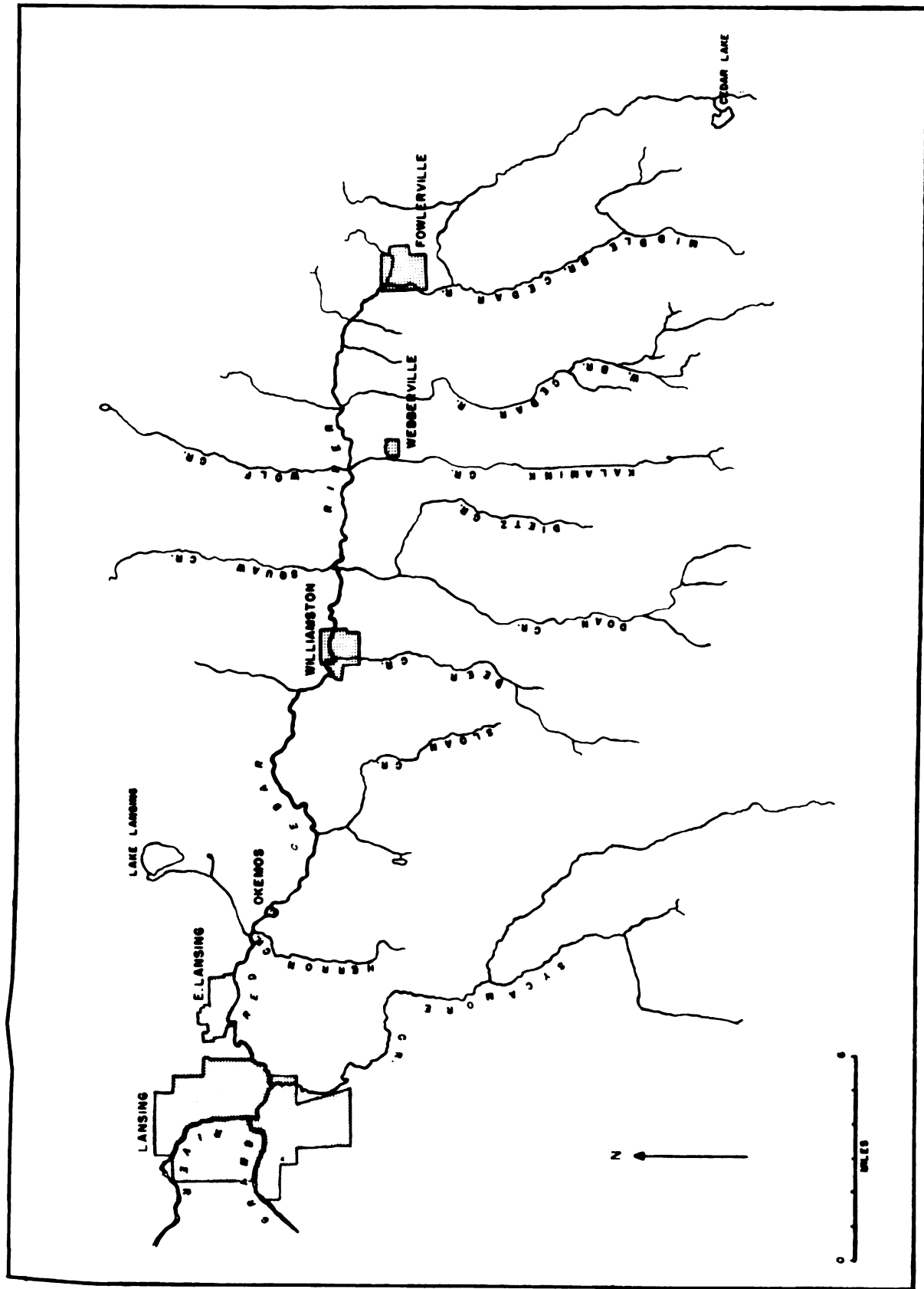


Figure 1.- Red Cedar River and Principal Tributaries

The original dam in Okemos was constructed to provide water power to operate a gristmill. The original structure has been replaced by a small stone ballast type that creates a small pool for recreational and aesthetic purposes. The East Lansing dam, located within the campus of Michigan State University, is constructed of concrete and maintains a constant depth of water for recreational purposes and for a cooling water supply for the steam-generated power plants on campus.

The bottom material of the Red Cedar River consists of fine sand in the upper regions to rocks and gravel with small areas containing silt deposition in the middle and lower stretches. In general, the bottom gradient is uniform with long pools or runs rarely exceeding four feet in depth divided by short riffle areas. The shoreline slope is gradual and covered with vegetation, resulting in very little erosion or cutting above the normal water line (Fig. 4).

The topography of the watershed area is nearly level to rolling as a result of the Cary phase of the Wisconsin glacier. The soils of the area are classified by Whiteside et al. (1956) as being derived from limy loam glacial till. The primary soil series are of the Miami and Conover types having good to intermediate drainage. A large proportion of the watershed area is used for dairy cattle grazing and small grain farming.

Physical and Chemical Characteristics

Two permanent stations are maintained on the Red Cedar River to record the physical characteristics of the stream. The first, a gauging station five miles upstream from the mouth, is maintained by the United

Fig. 4. Red Cedar River in the vicinity of Williamston, Michigan.



States Geological Survey to record the run-off for the 355 square mile area exclusive of the Sycamore Creek Drainage.

The flow is usually the greatest during March or April when the combination of melting snow, frozen ground, and spring rains occasionally result in floods that cause considerable property damage. The maximum discharge recorded at this station during the 1931-1958 period occurred in April 1947 when a flow of 5510 cubic feet per second was recorded. The minimum recorded during the above period was 3 cubic feet per second on July 31, 1931.

The median-mean monthly discharge for the 1947-1957 period and the rather atypical data for August 1957 through July 1958 are given in Figure 5.

The graduate students in Limnology, Department of Fisheries and Wildlife at Michigan State University maintain a temperature recording station at Dobie Road, 10 miles upstream from the mouth of the river. The daily high and low water temperatures for the period from July 1, 1957 to July 31, 1958 are given in Appendix A.

Twelve sampling stations were chosen along the main channel of the Red Cedar River within the area from 4.5 to 34.7 miles from the mouth to study the general physical and chemical characteristics of the water. Twelve series of samples were taken during June, July, August, and September 1956 and the following data were collected:

- | | |
|--------------------------|--------------------------------------|
| 1. Air temperature | 7. Dissolved oxygen |
| 2. Water temperature | 8. Total phosphorus |
| 3. Specific conductivity | 9. Nitrogen as ammonia |
| 4. Alkalinity | 10. Nitrogen as nitrite plus nitrate |
| 5. pH | 11. Gauge height at Mile 5 |
| 6. Turbidity | 12. Discharge at Mile 5. |

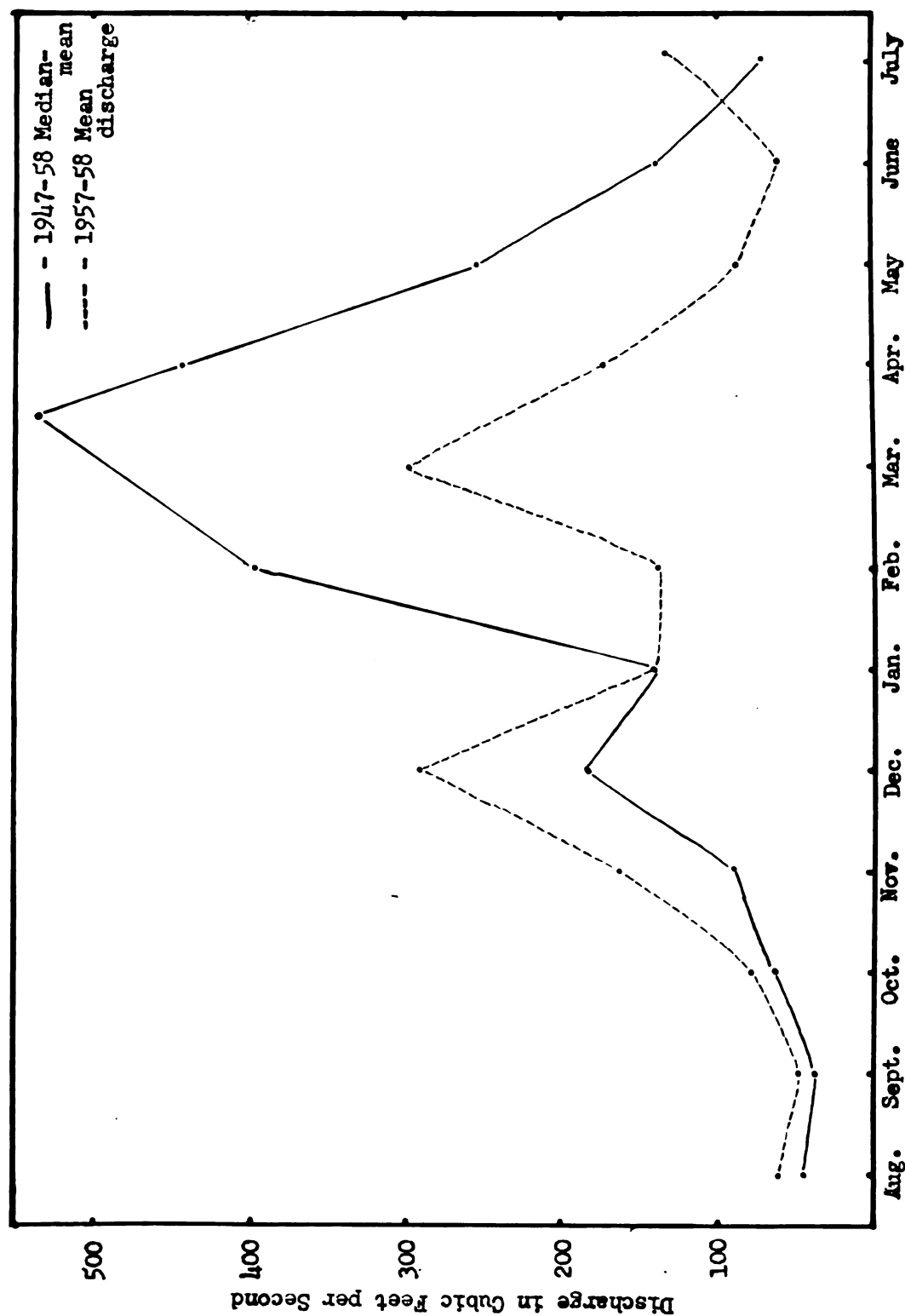


Fig. 5. Median-mean monthly discharge of the Red Cedar River for 1947-1958 and mean monthly discharge for the August 1957 through July 1958 study period. (Data from U.S. Geological Survey)

The values for this series of samples are given in Appendix B.

The specific locations of the twelve stations are given in Table 2.

It will be noted from the overlay for Figure 1 that the station sites were selected to make possible the measuring of nutrients from either tributary streams or municipal sewer outfalls.

The waters of the Red Cedar River are highly buffered and show little variation in pH. The bicarbonate ion content (expressed as CaCO_3) ranges from 250 to 300 mg l^{-1} and fluctuates inversely with stream flow. The pH values ranged from 7.4 to 8.3 during the initial survey period.

The high bicarbonate ion content contributes to the high specific conductivity of the stream. Values ranging from 372 to 590 $\times 10^{-4}$ ohms $^{-1}$ cm^{-1} were measured during the 1956 period with the wide range due to variations in water levels. An increase in the specific conductivity was noted toward the downstream areas. In view of the uniformity of the methyl orange alkalinity values between stations, the increase in specific conductivity as the water mass moves downstream can be attributed to the addition of other soluble salts.

The greatest change in chemical characteristics was detected in the basic nutrient elements, phosphorus and nitrogen (Figs. 6-10).

The data indicate that municipal effluents are responsible for the greater portion of the nutrients introduced into the stream and that the tributary streams contribute very little to the nutrient budget. Since Lund (1950) found that the aquatic flora can utilize phosphorus in concentrations as low as one microgram per liter, this element might

TABLE 2. Location of sampling stations used to determine the physical and chemical characteristics of the Red Cedar River.

<u>Station</u>	<u>Miles from Mouth</u>	<u>Location</u>
I	4.5	Located on the Michigan State University Campus opposite Stadium parking lot
II	7.8	Upstream from Indian Hilla Bridge in Okemos
III	10.3	Upstream from Dobie Road Bridge
IV	13.6	Upstream from U. S. Highway 16 Bridge
V	16.6	Upstream from Zimmer Road Bridge
VI	18.3	Off McCormick Street in Williamston
VII	18.8	0.3 mile downstream from Williamston Dam
VIII	23.5	Upstream from Michigan Highway 47 Bridge
IX	27.1	Upstream from Gramer Road Bridge
X	31.3	Upstream from Gregory Road Bridge
XI	34.3	Upstream from Van Buren Road Bridge
XII	34.7	Upstream from Fowlerville Road Bridge

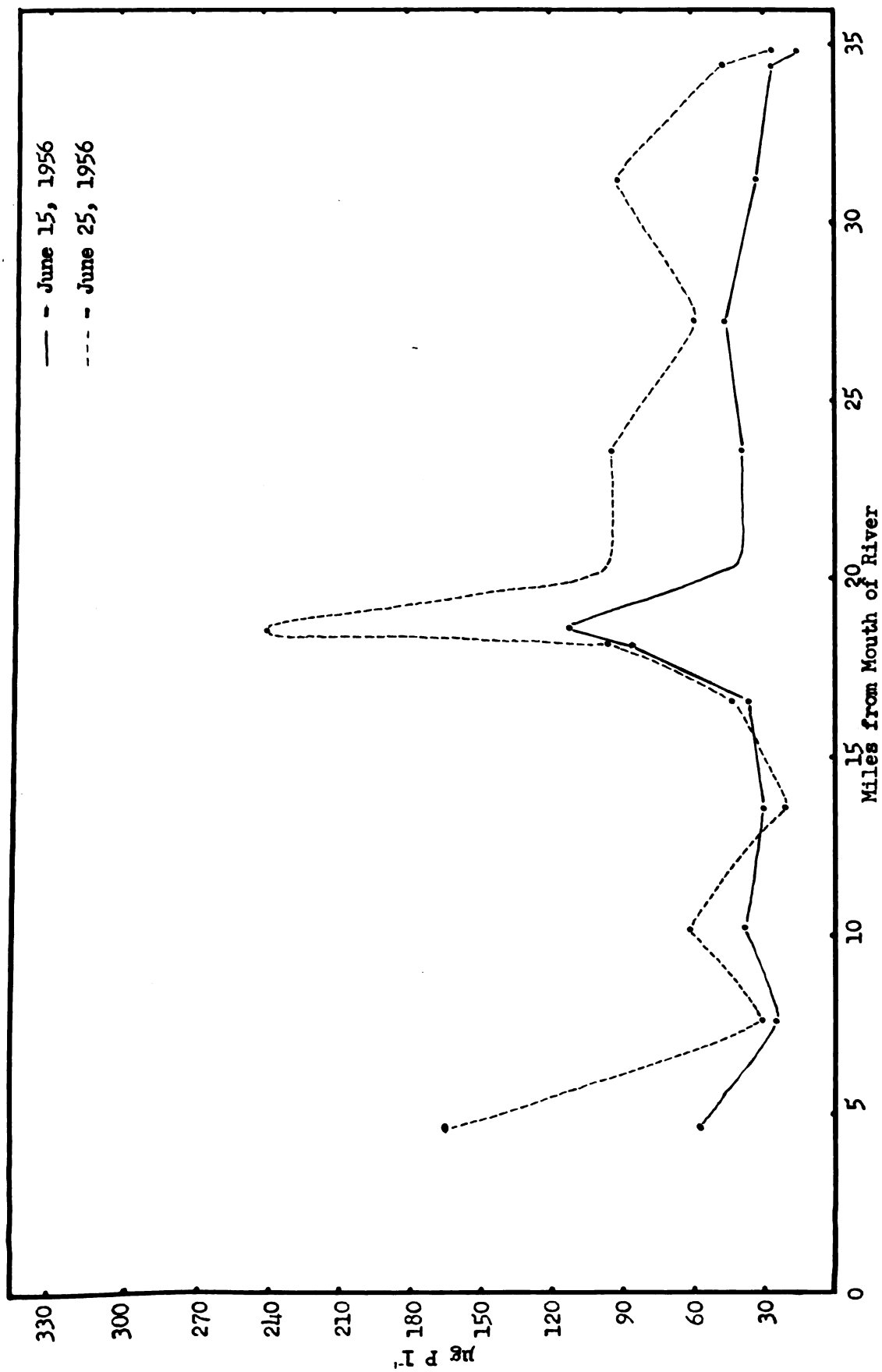


Fig. 6. Total phosphorus values for two sampling dates from 12 stations on the Red Cedar River.

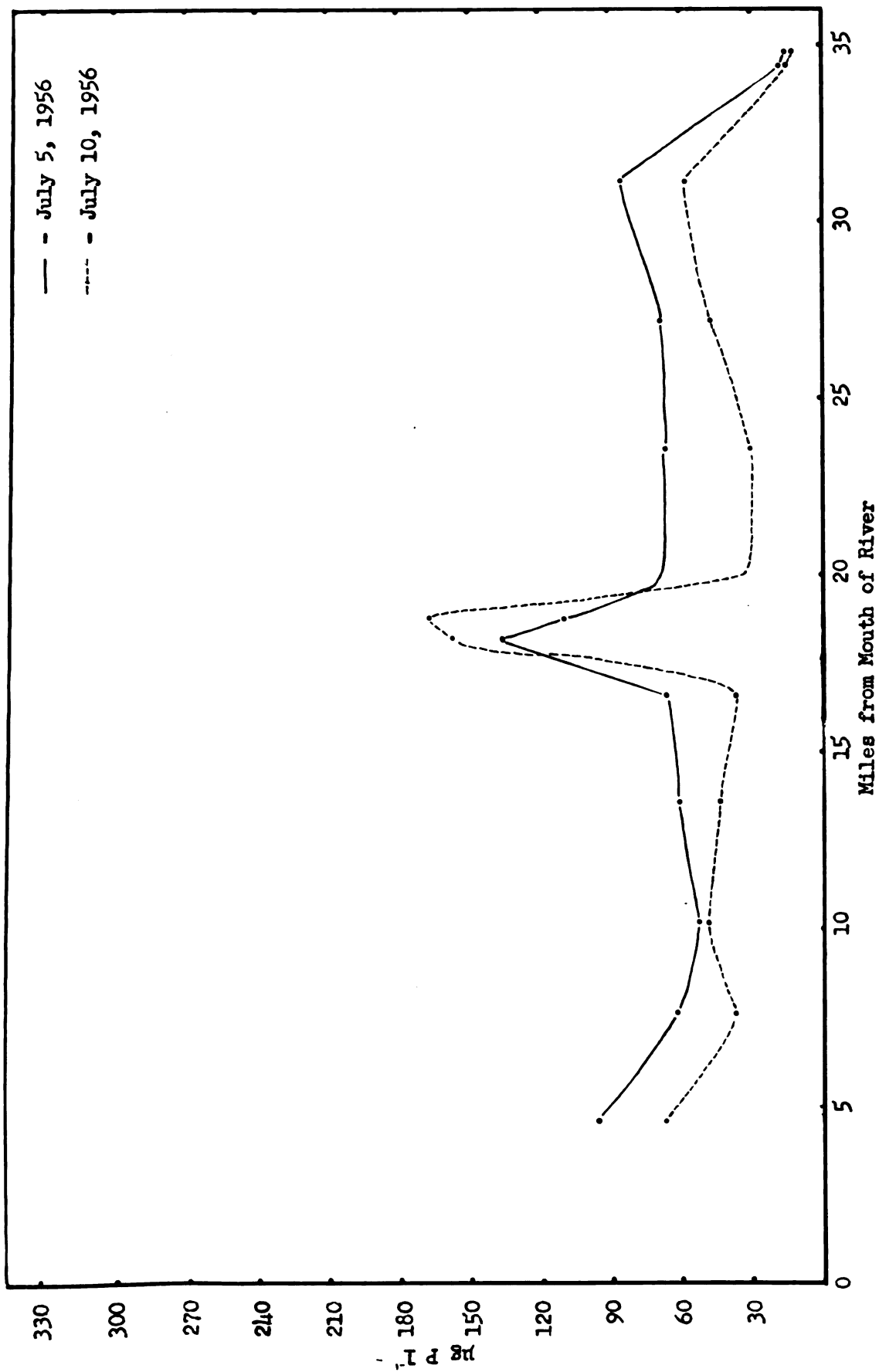


Fig. 7. Total phosphorus values for two sampling dates from 12 stations on the Red Cedar River.

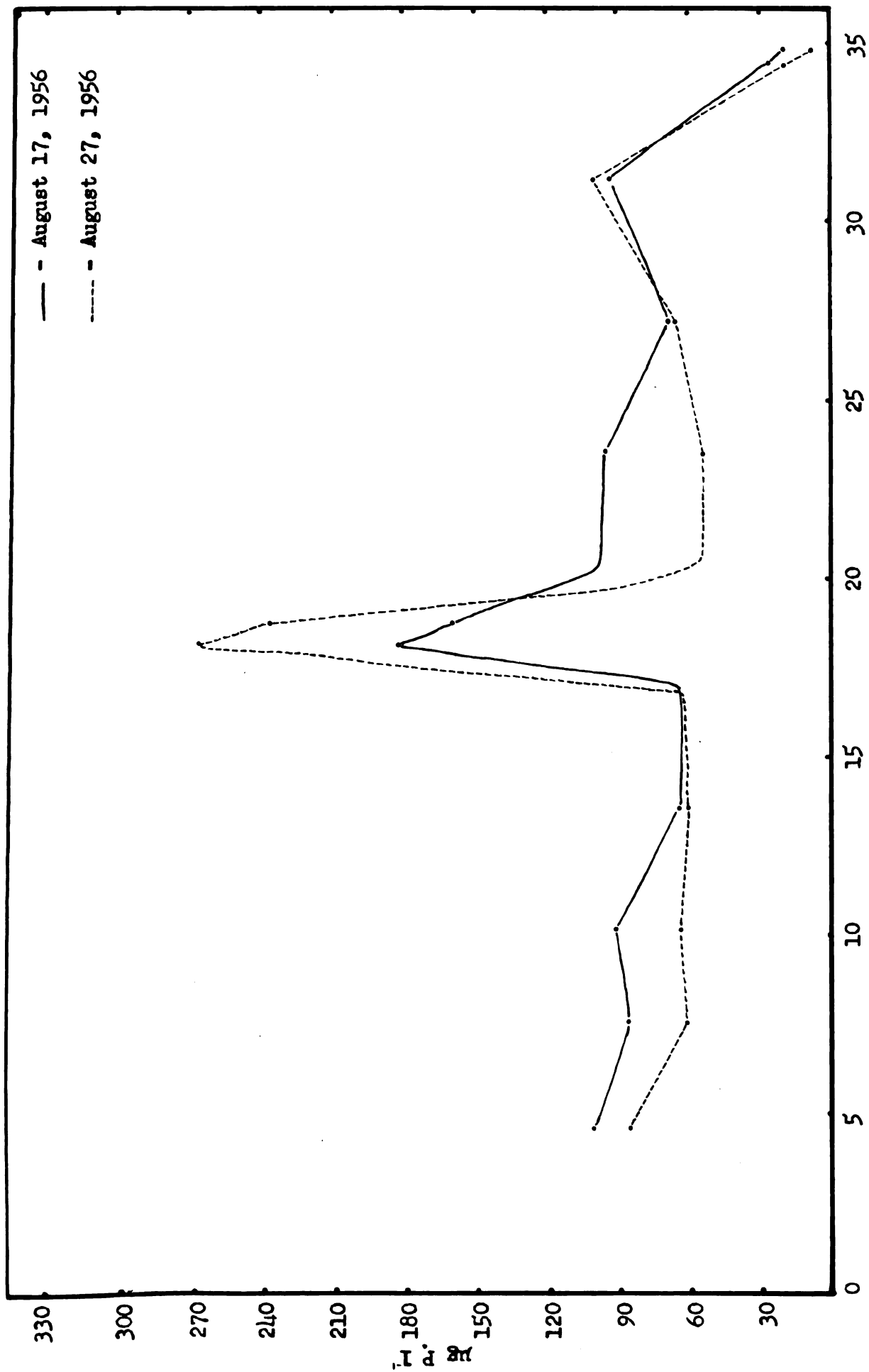


Fig. 8. Total phosphorus values for two sampling dates from 12 stations on the Red Cedar River.

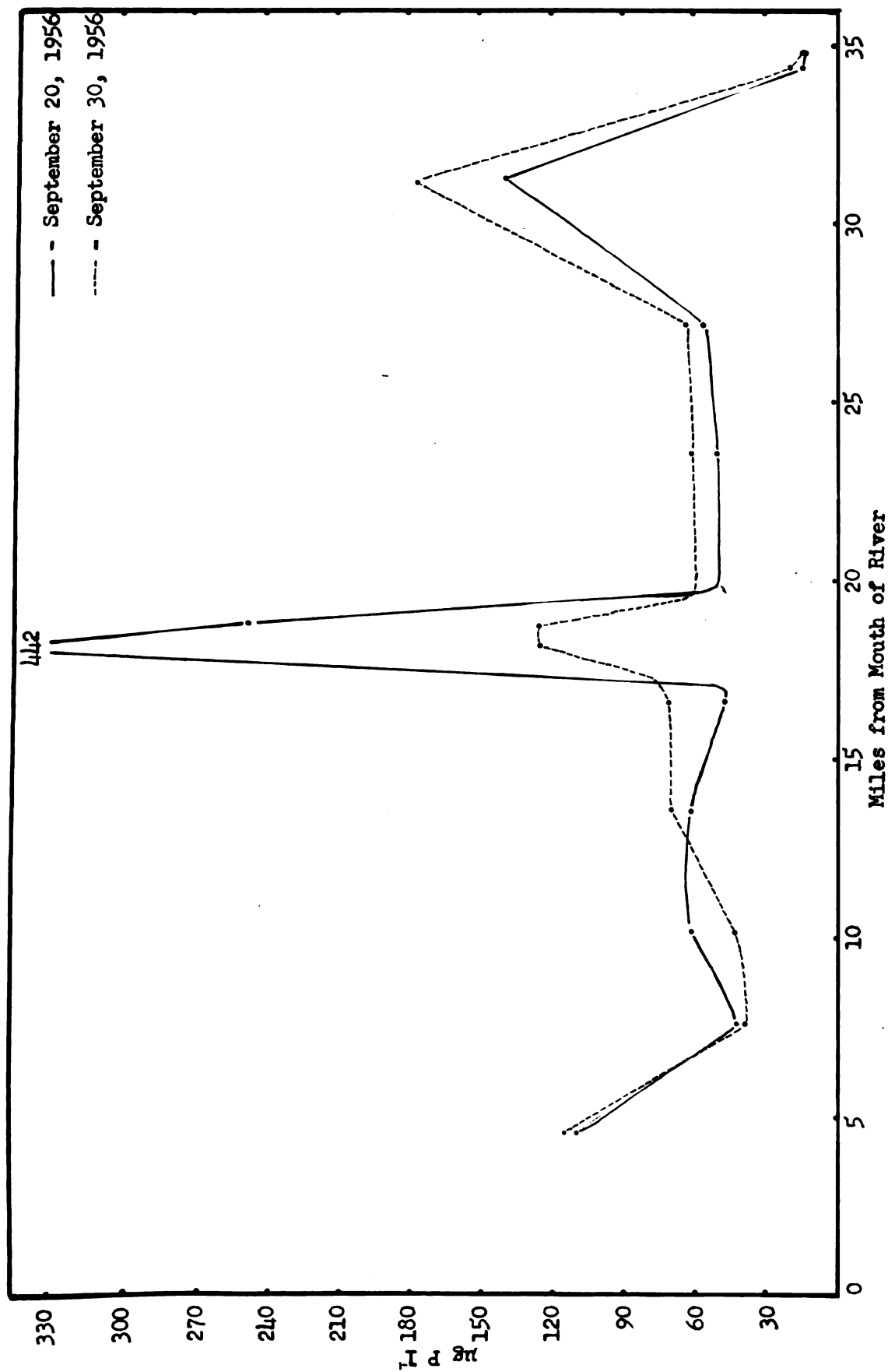


Fig. 9. Total phosphorus values for two sampling dates from 12 stations on the Red Cedar River.

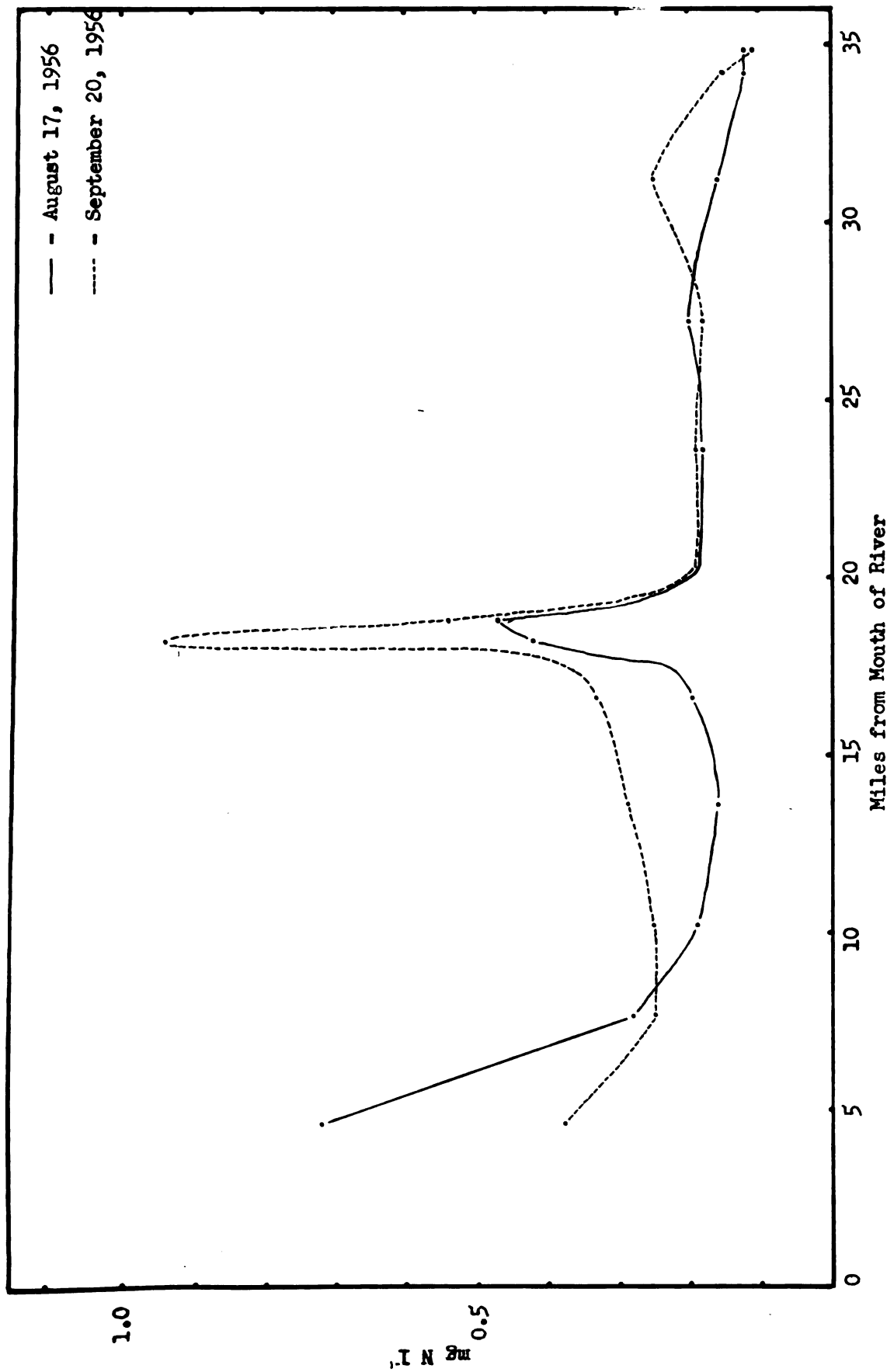


Fig. 10. Total inorganic nitrogen values for two sampling dates from 12 stations on the Red Cedar River.

always be considered as being present in the Red Cedar River at above the minimal level. The available nitrogen concentration was found to be relatively low in certain areas and may be a limiting nutrient for primary production in the Red Cedar River.

The greatest accrual of nutrients occurs within the Williamston City limits. During the initial survey period in the summer of 1956, before the sewage treatment plant was put into operation, nine tiles with an estimated total discharge of 100 gallons per minute fed waste water, raw sewage, and septic tank effluents into the stream. As the interceptor system was completed, the flow was diverted from these tiles to the sewage treatment plant. The effect of the diversion was apparent in the total phosphorus values as plotted in Figures 6 through 9.

In order to study the bio- and abiodynamics of the stream in the vicinity of the Williamston sewage treatment plant outfall, seven permanent and two temporary stations were established within the area from one-half mile above to 4.9 miles below the outfall. These stations, described by the distance in miles above or below the outfall¹, were designated as -0.4, -0.2, +0.1, +0.3, +0.6, +1.3, +1.8, +3.7, and +4.9 (Fig. 11). During the period from July 1, 1957 to August 15, 1958, the study was confined to this 5.3 mile area above and below the source of nutrients. Two stations were maintained upstream from the outfall. This was necessary to determine the chemical and biological effects of the Deer Creek drainage on the Red Cedar River. Thirty sets of water samples were taken for chemical analyses during the above period to study the relative enrichment, uptake, and regeneration of the nutrient elements and the periphyton production was measured for 24 periods by the phyto-pigment method previously described.

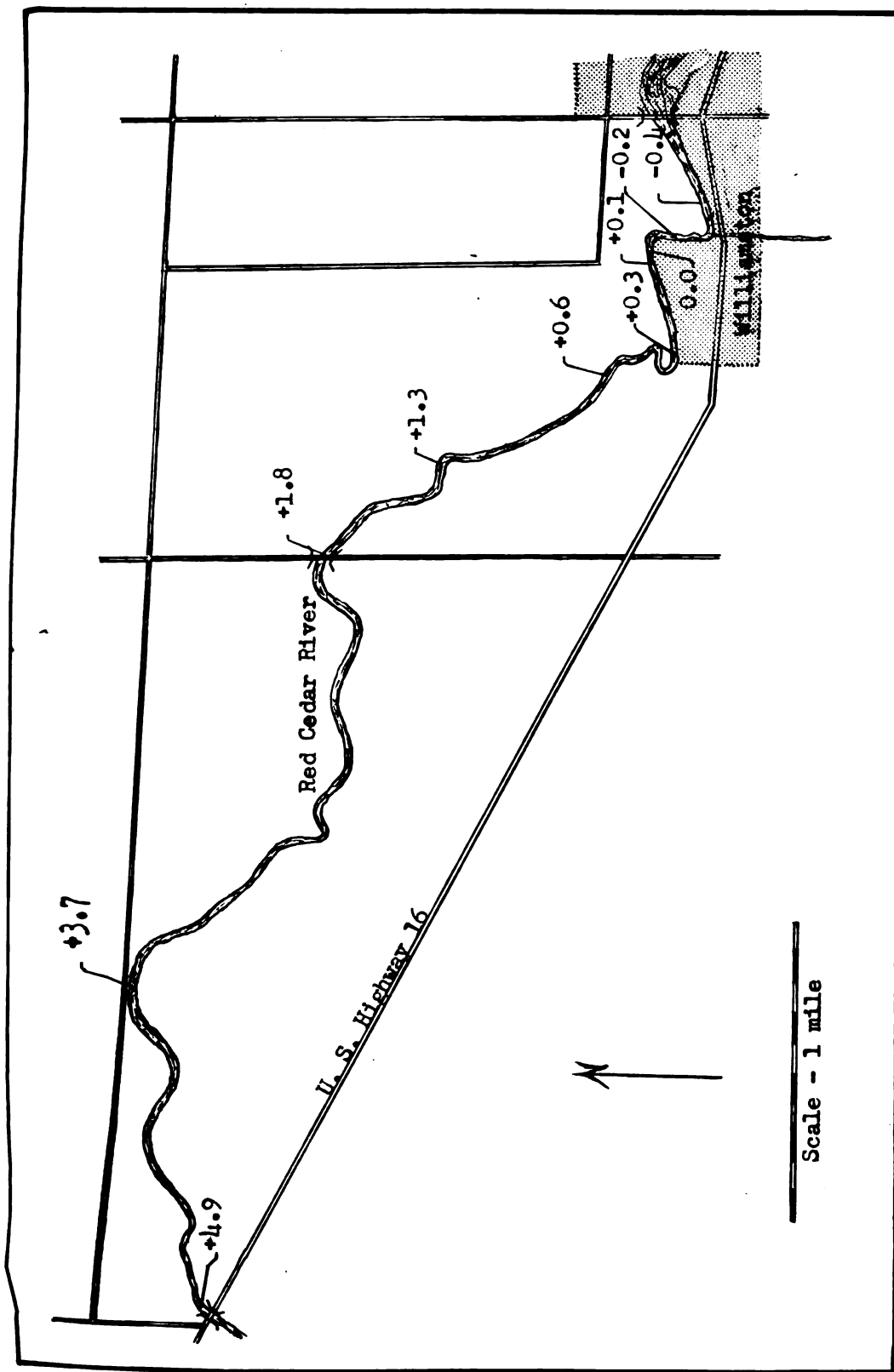


Fig. 11. Red Cedar River and the sampling stations which are numbered according to their distance (in miles) from the sewage treatment plant outfall.

RESULTS AND DISCUSSION

RESULTS AND DISCUSSION

Nutrient Metabolism

The mode of fertilization in this study differs from the artificial enrichment of a stream as reported by Huntsman (1948) in that the nutrient supply is in the form of sewage treatment plant effluent and is continuously supplied to the stream. This may result in the flora immediately downstream from the outfall becoming physiologically adjusted to high nutrient levels. The absence of toxic industrial wastes in the effluent and the inexistence of a septic zone in the stream should prevent the modification of the biocoenose and limitation of species as reported by Butcher (1947) or Blum (1957). Theoretically, the flora in the stream immediately downstream from the outfall would remain biologically "healthy" and capable of carrying on the normal metabolic functions.

Phosphorus

The study of the physical and chemical characteristics of the Red Cedar River indicated that municipal drains and sewage treatment plant outfalls are the most important sources of phosphorus. Although the tributary stream introduced water of high phosphorus content during periods of high flow, the adverse physical conditions which accompany the influx of nutrients prevent utilization and biological response by the flora of the stream.

The Williamston sewage treatment plant has a mean base effluent output of approximately 150,000 gallons per day. Since the interceptor system is a combination storm and sanitary type, the output and dilution vary with the local precipitation and run-off. Total phosphorus determinations were made on effluent samples collected by plant personnel on October 7 and 10, 1957 to determine the total phosphorus content of the effluent after it had passed through the primary treatment plant. No precipitation occurred during this period and the effluent discharge was not diluted by storm water. The results indicate a high variation in phosphorus content depending upon the time of the day and the day of the week. The 8 a.m. samples contained 7.1 and 7.8 mg P l⁻¹ respectively on the two dates. The highest value (23 mg P l⁻¹) was found in the sample taken at 2 p.m. on Monday, October 7. The frequency of occurrence of each phosphorus level in the effluent was estimated for the entire week. The daily, weekly, and annual phosphorus discharge with the effluent into the Red Cedar River was then calculated from the mean phosphorus value.

The mean total phosphorus output per day was computed to be approximately 6,800,000 milligrams. This is equivalent to 2.48 metric tons per year or an annual per capita phosphorus output of 8.5×10^{-4} metric tons. Although Rudolfs (1947) gives the phosphorus output per capita in sewage effluent after primary treatment as 2.21×10^{-4} metric tons per year, the calculated value for Williamston does not seem unreasonable since the sewage is more concentrated due to the absence of heavy industrial water users in the city and the increased use of household detergents which are high in phosphorus.

The level of phosphorus acquired by the Red Cedar River as the result of receiving the sewage treatment plant effluent varies with stream flow; but when the flow at Williamston is less than 50 cubic feet per second, it exceeds $100 \mu\text{g l}^{-1}$. Approximately two-thirds of the total phosphorus discharged into the river is in the dissolved form, but it may be combined in many complex inorganic and organic compounds. Although many complex forms of phosphorus are not immediately available to the aquatic flora, they may undergo hydrolysis or dephosphorulation as the result of bacterial action and be converted to an available form (Harvey, 1940).

The results of the chemical analyses indicate that nearly all of the dissolved phosphorus introduced by the effluent of the sewage treatment plant are removed from solution within the first 0.6 mile below the outfall. In order to determine the rate of removal, midstream water samples were taken during a low water period on August 25, 1957 at locations 50, 100, 200, 300, 400, 500, 600, and 700 yards downstream from the outfall and the total dissolved and total sestonic phosphorus content determined. The results, along with those obtained August 20, 1957 for upstream stations -0.2 and -0.4, are given in Table 3.

Even though the effluent is not completely mixed with the river water after flowing 50 yards downstream from the outfall and after passing through a short riffle area, the reduction in the dissolved phosphorus far exceeds the theoretical reduction calculated for dilution. This indicates that phosphorus is being removed from solution either by chemical precipitation or biological uptake.

Abiotic phosphorus removal. In a highly buffered stream having a high pH one might theorize that the soluble phosphorus combines with the

TABLE 3. Dissolved and sestonic phosphorus values of water samples taken August 20 and August 25, 1957 from stations located above and below the sewage treatment plant outfall.

<u>Sampling location</u>	<u>Dissolved phosphorus</u>	<u>Sestonic phosphorus</u>
0.4 mile upstream ¹	51 $\mu\text{g l}^{-1}$	72 $\mu\text{g l}^{-1}$
0.2 mile upstream ¹	47	49
50 yards downstream	130	101
100 yards downstream	45	63
200 yards downstream	45	62
300 yards downstream	50	73
400 yards downstream	39	66
500 yards downstream	50	64
600 yards downstream	54	73
700 yards downstream	46	61

¹Samples taken August 20, 1957.

calcium to form one of the insoluble complexes and be precipitated from solution. The relationship between phosphorus loss and alkalinity of the water in a lentic environment has been described by Barrett (1952).

In a stream characterized by continual agitation the inorganic crystals of a calcium-phosphate complex would remain in suspension for some distance downstream from the point of formation. Upon filtration of a water sample, these crystals would be retained on a Millipore filter pad and, after being removed by a dilute acid solution, could be measured quantitatively by the method previously described.

The acid-soluble sestonic phosphorus was determined for seven periods during the period from July 1, 1957 to July 31, 1958. The results are given in Table 4.

TABLE 4. Micrograms of acid-soluble sestonic phosphorus in 100 ml water samples from nine stations on the Red Cedar River.

Date	<u>Station</u>								
	<u>-0.4</u>	<u>-0.2</u>	<u>+0.1</u>	<u>+0.3</u>	<u>+0.6</u>	<u>+1.3</u>	<u>+1.8</u>	<u>+3.8</u>	<u>+4.9</u>
May 15, 1958	4.3		3.2	2.6	1.6	1.6	1.7	1.7	1.7
27	3.0		7.8	2.4	2.1	2.2	2.2	1.3	1.3
June 2	3.3	3.1					2.2		
19	3.1	1.9	5.4	2.4	2.2	2.4	2.3	1.2	1.2
July 1	2.6	2.2	7.6	2.3	2.3		2.5	1.7	1.9
23	2.4	1.9	5.4	1.9	1.9		2.0	1.4	3.0
31	3.1	1.9	5.6	2.0	2.0		1.9	1.0	1.3

The results of the analyses indicate an increase in the acid-soluble fraction at Station +0.1, but the values at Station +0.3 were comparable to those found for the stations located upstream from the outfall. The data would appear to indicate that although a small amount of acid-soluble inorganic phosphorus is in suspension immediately below the outfall the reduction in soluble phosphorus cannot be attributed to precipitation.

A large amount of euplankton flows over the dam from the reservoir upstream from Station -0.4 during the summer months and the amount of plant material in the water as indicated by the green color of the filter pads shows a gradient from the upstream to the downstream stations. With the exception of the slight increase at Station +0.1, the reduction of acid-soluble phosphorus appears to be closely related to the decrease in plankton in the water. Apparently a portion of the phosphorus in the decomposing limnoplankton is acid-labile after the cells fail to survive the rigors of the lotic environment.

Biotic phosphorus removal. Having failed to demonstrate an abiotic mechanism responsible for the reduction in phosphorus in the area immediately downstream from the outfall, the flora of the stream was investigated.

Although the rooted aquatic plants and trees along the shore undoubtedly obtain a portion of their nutrients from the river, the periphyton of the stream is in contact with a greater part of the water mass and would be more likely to be responsible for phosphorus removal.

The quantitative periphyton study described later indicated that even when the increase in periphyton production occurred in the areas

several miles from the outfall, the decrease in dissolved phosphorus between Stations +0.1 and +0.6 remained relatively constant. If the periphyton in the enriched area was storing phosphorus to levels above its normal requirements as found by Einsele (1941), the mechanism for the removal of the element from solution even during periods when the periphyton production was low could be detected by determining the phosphorus:weight ratio of the growth on the substrata from the different stations.

In order to determine the phosphorus:weight ratio of the periphyton, total phosphorus determinations were made on the growth on one substratum from each set at each station except +0.1. The latter was eliminated from this portion of the study because the results might be erroneous due to nutrient-rich particulate material filtered from the water by the periphyton growth. The periphyton on the remainder of the substrata from each station was treated to determine the relative amount of growth. The results are given in Table 5.

TABLE 5. Micrograms of organic phosphorus:relative periphyton growth ratio as determined at seven stations on the Red Cedar River.

<u>Station</u>	<u>µg P/phytopigment unit</u>
-0.4	0.72
-0.2	0.57
+0.3	2.50
+0.6	1.62
+1.8	0.97
+3.8	0.75
+4.9	0.64

The ratio of phosphorus to organic material in periphyton increases greatly after the introduction of the nutrient-rich effluent and then decreases toward Station +4.9. It is interesting to note that the decrease in the phosphorus:organic material ratio between Stations -0.4 and -0.2 follows a decrease in the dissolved phosphorus in the water (Fig. 12), but after 0.6 mile below the outfall the dissolved phosphorus in the water increases but the phosphorus:organic matter ratio continues to decrease. Although the data for the stations downstream from Station +0.6 may appear anomalous due to the increase in dissolved phosphorus and decrease in the phosphorus content of the periphyton, it should be pointed out that the values derived from a chemical analyses of the water are a measurement of the elements not utilized by the flora of the stream. Also, a dynamic population of periphyton does not tend to accumulate phosphorus above the needs of the organisms, but instead utilizes the element in the production of new cells (Grzenda, unpublished). It may be assumed then that the greater portion of the phosphorus introduced with the effluent into the Red Cedar River is biologically removed from solution within a very short distance from the sewage treatment plant outfall.

Phosphorus regeneration. The increase in dissolved phosphorus downstream from Station +0.6 (Fig. 12) raises a question concerning the mechanism of phosphorus transport from the area of "high" concentration through a distinct area of "low" concentration where the dissolved phosphorus values may at times be nearly equal to those upstream from the effluent discharge to the downstream areas where the values increase with distance from the outfall. One might assume that the downstream

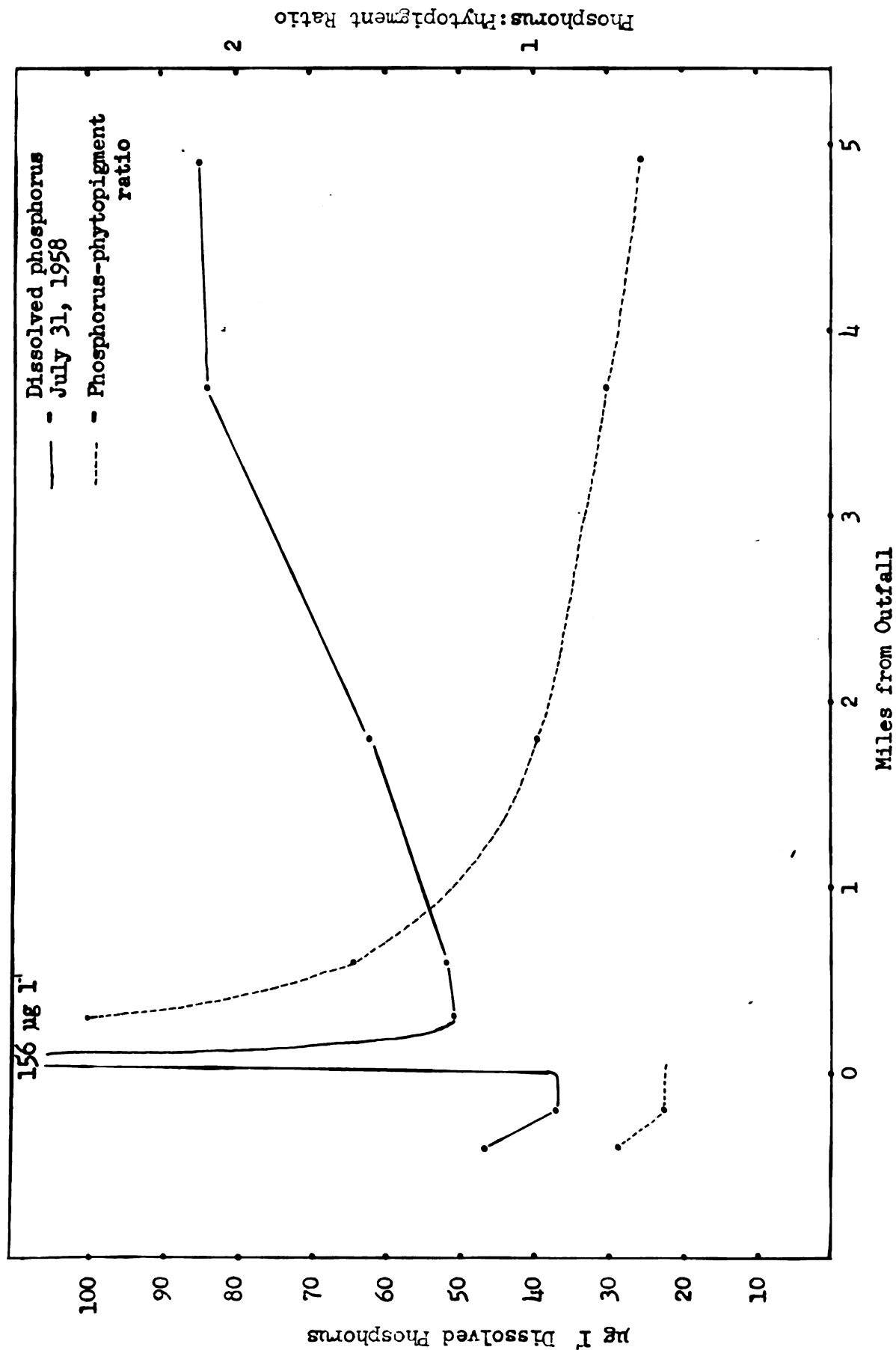


Fig. 12. Dissolved phosphorus content of the water and the phosphorus:phytoplankton ratios of collected periphyton from stations on the Red Cedar River for the July 29 to August 5, 1958 period.

increase was due to the mineralization of particulate organic waste released in the effluent and that the area of stabilization would move upstream as the water temperature increased and downstream as the water temperature decreased. Although the zone of phosphorus regeneration tends to shift with water temperature, the method of transport cannot be associated with suspended solids. The total sestonic phosphorus values for the station 0.3 mile below the outfall were usually comparable to those upstream from the outfall. Therefore, it must be assumed that the phosphorus is transported biologically from the areas of "high" concentration to the zone of regeneration.

It was demonstrated that the periphyton 0.3 mile downstream from the source of nutrients contained over four times as much organic phosphorus per unit of growth as the periphyton upstream from the outfall. One can also assume that this ratio increases between Station +0.3 and the outfall. The quantitative periphyton data indicate that the net amount of organic material produced may exceed $20 \text{ mg dm}^{-2} \text{ day}^{-1}$ in the more productive areas of the Red Cedar River (Grzenda, unpublished). If it were not for a relatively short turnover period, i.e. sloughing and repopulation of an area, the stream bed would soon be covered with a thick mat of periphyton. The sloughing process results when new growth excludes the light from the basal cells causing them to die and become detached from the substratum. The disintegration of the basal cells may be accelerated by the nascent oxygen produced by the outer photosynthetic layer whose growth in turn may be stimulated by the carbon dioxide and nutrients released from the basal layer.

The downstream increase in dissolved phosphorus results when the periphyton containing the high phosphorus:weight ratio becomes detached

from the substratum, is carried downstream by the current, disintegrates and undergoes decomposition, and releases the nutrients to solution. The area of nutrient regeneration is governed by the stream velocity and the water temperature.

The role of water temperature in the decomposition rate of the detached periphyton is well demonstrated in Table 6. On August 19, 1957 and June 19, 1958 when the daily high for the water temperature approached 20° C., the sestonic phosphorus content of the water was much lower at Station +3.7 than at Station +0.6. On these dates the regeneration was most apparent as shown in Figure 13. On January 14, 1958 and April 15, 1958 when the water temperature was below the optimum for bacterial activity, the sestonic phosphorus values were equal for the two stations.

TABLE 6. Sestonic phosphorus content of the water at two stations on the Red Cedar River as related to water temperature.

<u>Date</u>	<u>Water temperature</u>		<u>Station</u>	
	<u>Low</u>	<u>High</u>	<u>+0.6</u>	<u>+3.7</u>
August 19, 1957	17.9° C	20.6° C	59 µg l'	26 µg l'
January 14, 1958	0.0° C	0.0° C	26 µg l'	26 µg l'
April 15, 1958	8.8° C	13.4° C	33 µg l'	33 µg l'
June 19, 1958	16.8° C	19.4° C	52 µg l'	28 µg l'

During the winter months when bacterial activity is greatly reduced, the area of greatest phosphorus regeneration was downstream from the study area. Mr. Alfred R. Grzenda, a fellow graduate student who was studying the biodynamics of the stream at an area 9.9 miles downstream from the outfall, consistently obtained higher values than were obtained

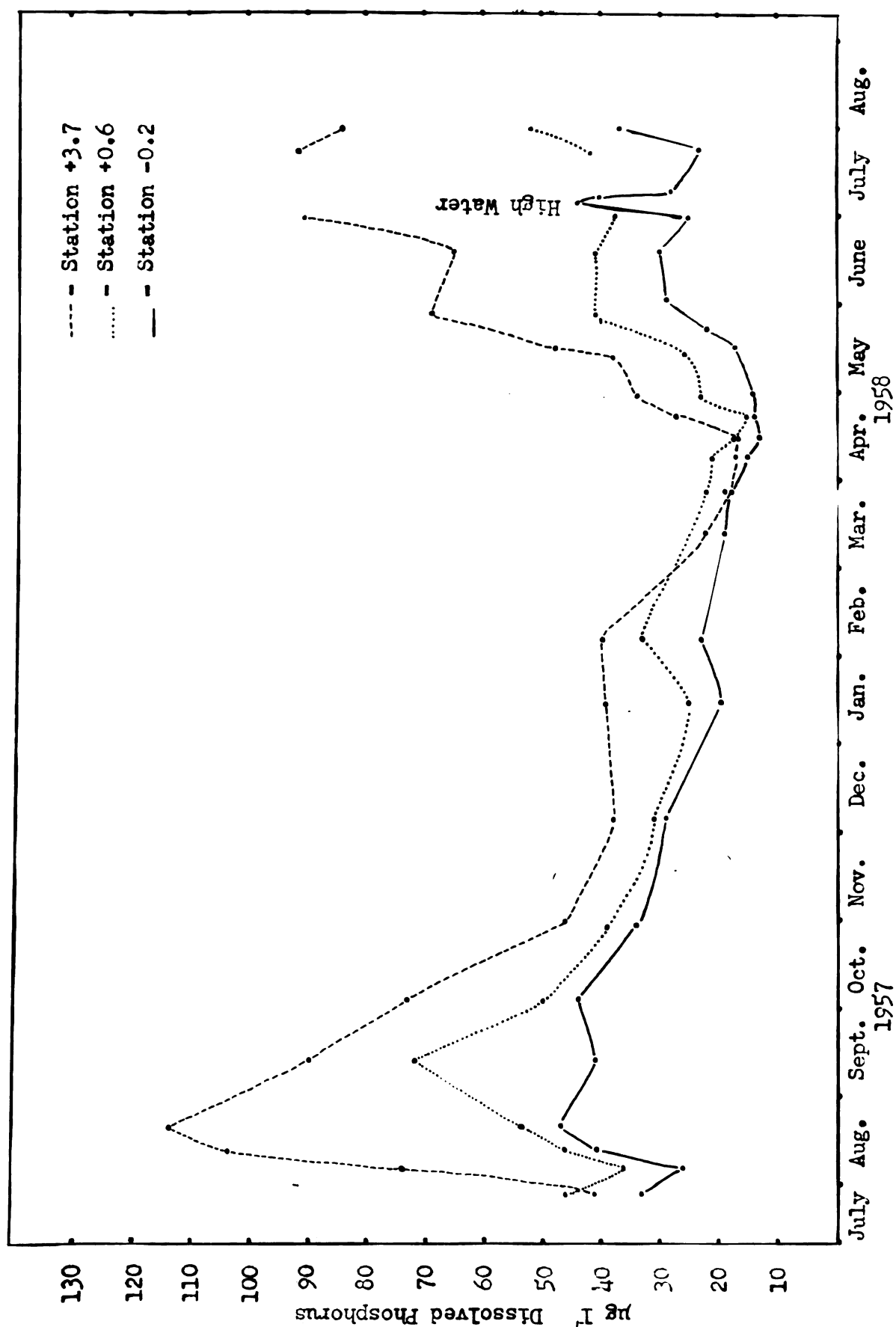


Fig. 13. Micrograms of dissolved phosphorus per liter of water as determined at three stations on the Red Cedar River.

at Station +4.9. After the end of the period of ice cover (March 3) the periphyton growth was stimulated by the additional light reaching the substratum and the dissolved phosphorus content of the water was reduced in the downstream areas. After the water temperature had increased sufficiently to stimulate bacterial activity, the zone of regeneration again shifted upstream to within the 5.3 mile study area and the phosphorus accrual exceeded biological utilization. This resulted in an accumulation of the element in the dissolved form and produced the downstream regeneration curve as shown in Figure 12.

Ammonia Nitrogen

Although ammonia nitrogen is seldom a constituent of streams, it was of great importance in this study because ammonia is the first inorganic nitrogen form produced in the nitrification of organic material and is usually present in sewage treatment plant effluent. Also, the reservoir located above Station -0.4 contained large amounts of plant material and organic depositions which proved to be a source of ammonia for the section of stream under investigation.

In streams that do not receive organic wastes the ammonia produced is almost immediately utilized by the aquatic flora or converted to nitrites and nitrates. The latter is accomplished through bacterial action. The ammonia present in water is mainly in the form of NH_4^+ and undissociated NaOH . At the normal summer pH values of the Red Cedar River the ratio between NH_4^+ and NaOH would be approximately 30:1.

The amount of ammonia produced in the Williamston sewage treatment plant and emptied into the Red Cedar River with the effluent is high for this type of installation. This might be explained in part by

the plant's operation at only 60 percent of designed capacity resulting in a longer retention period of the liquid fraction in the primary settling tank. Also, the sludge must be allowed to accumulate for a longer period of time to obtain an adequate amount to justify withdrawal and transfer to the composting unit. The ammonia nitrogen values obtained at Station +0.1 ranged from trace amounts during the July 1957 flood period to 1.2 mg l⁻¹ during the low water period in July of 1958. The effluent is not completely mixed with the river water at Station +0.1, and the ammonia nitrogen content of the water is reduced by dilution, conversion to nitrites and nitrates, and plant utilization within the next 0.2 mile so that the highest value received at Station +0.3 was 0.42 mg l⁻¹.

During the summer months when bacterial and periphyton activities were high, significant ammonia nitrogen values were recorded only at Stations +0.1, +0.3, and occasionally at +0.6. One exception to the above was noted during the study period and this was attributed to an industrial accident approximately 15 miles upstream from the area.

On May 21, 1958, a fire destroyed a portion of a metal plating factory located at Fowlerville, Michigan. In the course of fighting the fire, water flooded the cyanide tanks and this chemical was diverted into a waste treatment lagoon. The plant manager quickly ordered the lagoon outlet closed and initiated treatment of the lagoon contents with sodium hypochlorite to oxidize the cyanide to the relatively non-toxic cyanate (Eldridge, 1933; Dobson, 1947) and thus prevented what might have been a major catastrophe to the stream. When the reaction was complete, the contents of the lagoon were fed into the Red Cedar

River. Since cyanates are hydrolyzed to ammonia compounds, the rate of which is a function of the pH of the receiving water, it is theorized that this reaction accounted for the presence of ammonia at both the upstream and downstream stations within the study area during June 1958. Although Resnick et al. (1958) found that the cyanates were relatively stable under aerobic conditions, it is theorized that the formation of ammonia may result from microbial action in the complex natural environment.

During the fall and winter months when the biological activity in the area was low, ammonia nitrogen was found at all stations. The periphyton data indicate that when the phytopigment production fell below approximately five units per square decimeter per day the ammonia production exceeded utilization and conversion.

The nitrogen introduced into the Red Cedar River in the form of ammonia cannot be followed through the uptake and regeneration phases that were associated with the phosphorus. Even though the quantity of ammonia received by the stream is high during the summer period, it is unlikely that it would be lost to the atmosphere in the form of gas since 800 volumes can be absorbed in one volume of water at 20° C. Also, since the amount of nitrogen in the oxidized forms does not increase at Station +0.6, it must be assumed that during the summer period the ammonia nitrogen is assimilated into organic material within a short distance downstream from the outfall.

The nitrogen cycle in the study area will be covered more fully in the section discussing nitrates.

Nitrogen as Nitrite plus Nitrate

The nitrate ion, unlike the phosphate ion, is not firmly held by soil particles and is easily leached from the topsoil and eventually finds its way into lakes and streams. Schmidt (1956) in his work in Minnesota reports finding 6.4 mg l⁻¹ of nitrates in spring water from Nobles County and 18 mg l⁻¹ in water from a field tile draining cultivated land. In view of the above data one might expect a stream receiving not only drainage water from agricultural areas, but also sewage treatment plant effluent, to contain large amounts of nitrates. However, as Whipple (1948) points out, the nitrogen measured in a stream represents only that which has not been utilized by the aquatic flora. In a small stream system such as the Red Cedar River the flow in many of the tributary streams is less than one cubic foot of water per second except during periods of rainfall and the periphyton community and rooted vegetation of the tributary removes the excess nutrients from the water before it reaches the main stream. The relationship between the tributary waters and the nutrient content of the water in the study area could be detected only during periods of heavy run-off and during the winter months when the biological activity was low. The nutrient-rich water received from the tributary streams during periods of heavy rainfall also carries a heavy sediment load which, as will be demonstrated later, destroys the standing crop of periphyton. Therefore, the bulk of these nutrients cannot be utilized by the aquatic flora and are carried downstream without producing a biological response.

The highest $N \cdot NO_2 + NO_3$ value, 4.12 mg l⁻¹, was recorded on July 5, 1957 when the Red Cedar River was above flood stage. When the water

level receded, the nitrogen value dropped to 0.2 mg l^{-1} .

The month of July 1958 provided an excellent period to study the relationship between precipitation, stream flow, and oxidized nitrogen in solution in the river (Fig. 14). The stream flow and precipitation data were recorded at the East Lansing Stations by the U. S. Geological Survey and the U. S. Weather Bureau while the nitrogen values were determined on water samples taken at Station -0.2 of the study area.

The river was nearly at base flow prior to the rains on July 3, 4, and 5 which caused the water level to rise slightly over four feet at Station -0.2. Since the watershed area was very dry prior to the rains, the surface run-off period was very short and the river level dropped 23 inches between July 5 and July 7, and 13 inches between July 7 and July 9.

The $\text{N} \cdot \text{NO}_2 + \text{NO}_3$ in the river increased from 0.11 mg l^{-1} before the rainfall period to 2.70 mg l^{-1} during the peak run-off period. It then decreased rapidly as the run-off declined. The nitrogen value was only 0.29 mg l^{-1} on July 23 and 0.06 mg l^{-1} on July 31.

During the study period the $\text{N} \cdot \text{NO}_2 + \text{NO}_3$ content of the Red Cedar River did not show a significant increase after the introduction of the Williamston sewage treatment plant effluent even though as much as 1.2 mg l^{-1} of nitrogen in the form of ammonia was measured at Station +0.1 in June 1958. This would appear to indicate that even though nitrification may occur, the utilization rate of ammonia by the aquatic flora of the stream is sufficiently high to prevent accumulation of the oxidized forms.

One might question why nitrification and an accumulation of nitrogen in the oxidized forms do not occur during the winter months when the

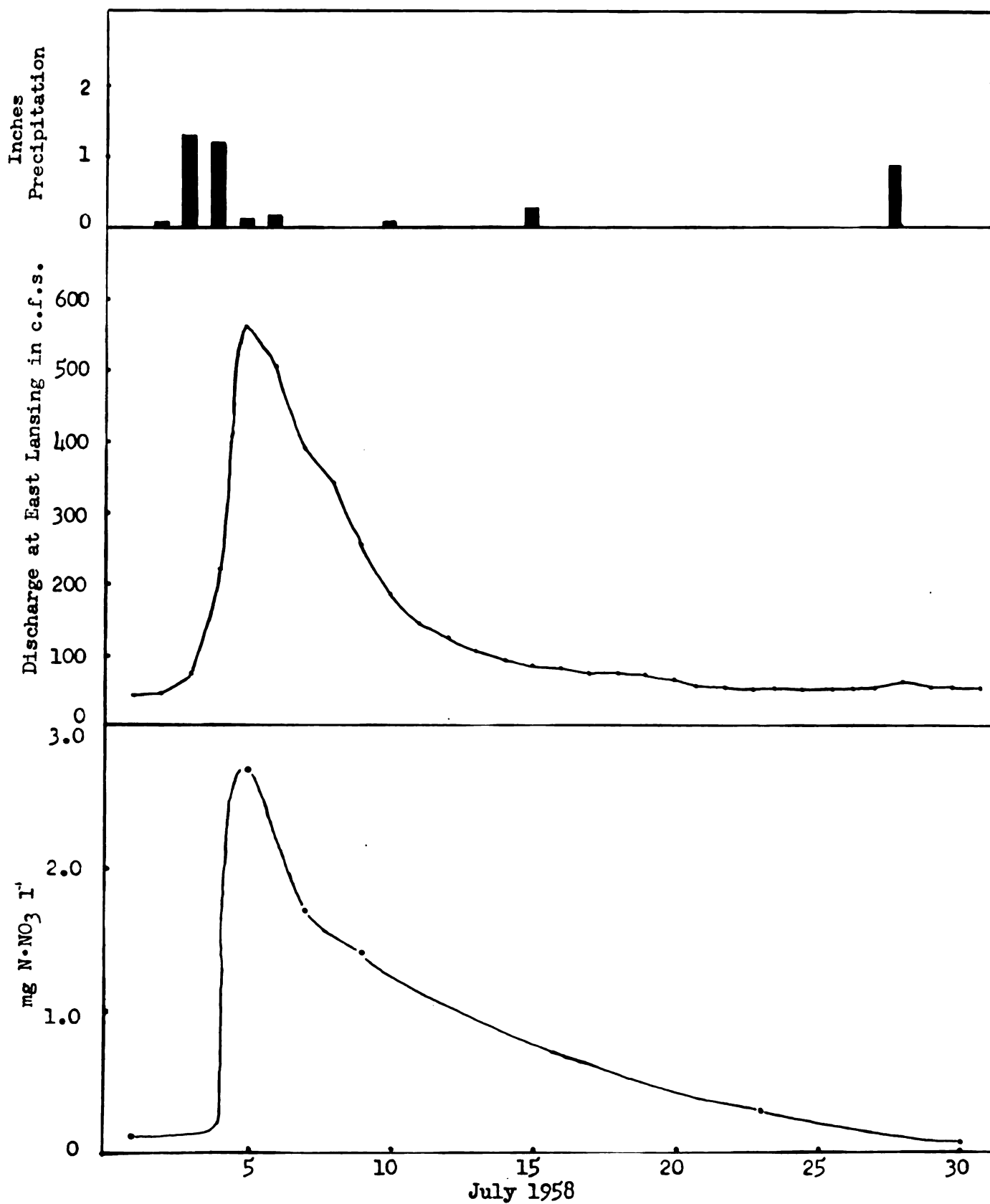


Fig. 14. Relationship between precipitation, stream flow, and inorganic nitrogen in the Red Cedar River.

periphyton production was low. First, the rate of mineralization of nitrogeous waste in the settling basin of the sewage treatment plant proceeds much more slowly during cold weather and the production and release of ammonia was less than 50 percent of the summer values. Second, the activity of the organisms responsible for the nitrification of ammonia would be reduced by the suboptimal temperatures so that the increase would occur further downstream from the outfall. And third, in a lotic environment the ammonia introduced is carried downstream as NH_4OH or as NH_4^+ either as the free ion or as the ion adsorbed on organic particles so it does not accumulate in a given area and undergo nitrification.

During the winter period the $\text{N}\cdot\text{NO}_2 + \text{NO}_3$ values were slightly higher at the downstream stations than at those located upstream from the sewage treatment plant. The build-up occurred when the downstream stations were covered with ice and the photosynthetic activity was low. The $\text{N}\cdot\text{NO}_2 + \text{NO}_3$ values at Station +3.7 were greater than 1.0 mg l^{-1} on the sampling dates between November 19, 1957 and March 26, 1958, but values of less than 0.5 mg l^{-1} were found after April 22, 1958.

The data indicate that the $\text{N}\cdot\text{NO}_2 + \text{NO}_3$ values in the Red Cedar River are highest during the winter months when the stream is covered with ice and periphyton production is low. After the spring thaw the periphyton production increases and the nitrogen content of the water decreases. During the summer months the available nitrogen content of the water may increase temporarily as the result of precipitation and run-off from the watershed area, but concentration in solution decreases sharply after the peak run-off period. During the periods of high

periphyton production the total inorganic nitrogen content of the water was frequently less than 0.1 mg l⁻¹.

Periphyton Production

The periphyton occupies a very important position in the energy transfer systems operating within a lotic environment. Because euplankton is virtually absent from all but the more sluggish streams, the role of converting solar energy to organic material is carried out almost entirely by these forms. Occasionally rooted plants such as Vallisneria sp. or Potamogeton sp. are found anchored to the stream bed, but these forms are usually of minor importance because of their seasonal occurrence, limited distribution, and inability to recover quickly from adverse conditions. The periphyton community is ubiquitous, and although seasonal variations in composition and productivity do occur, the quantities produced are sizeable at all times of the year because of its short turnover period. Besides its position as the most important primary producer in a stream, the periphyton mat also provides shelter for numerous forms of benthic organisms. The herbivores could be expected to be found in this "green pasture", but the fauna also includes various filter feeding forms and carnivores.

Although the 1.4 square decimeter plexiglass substrata used to measure the periphyton production in the Red Cedar River presented a smoother growing surface for the organisms than is naturally found in a stream, the growth patterns on the substrata appeared to be comparable to that on the stream bed. In other words the phytopigment density on the substrata was high when the periphyton population on natural submerged

objects appeared to be adding new growth. Likewise, when the flora on the natural substratum appeared to be in a static state, the production as measured on the plexiglass plates was low.

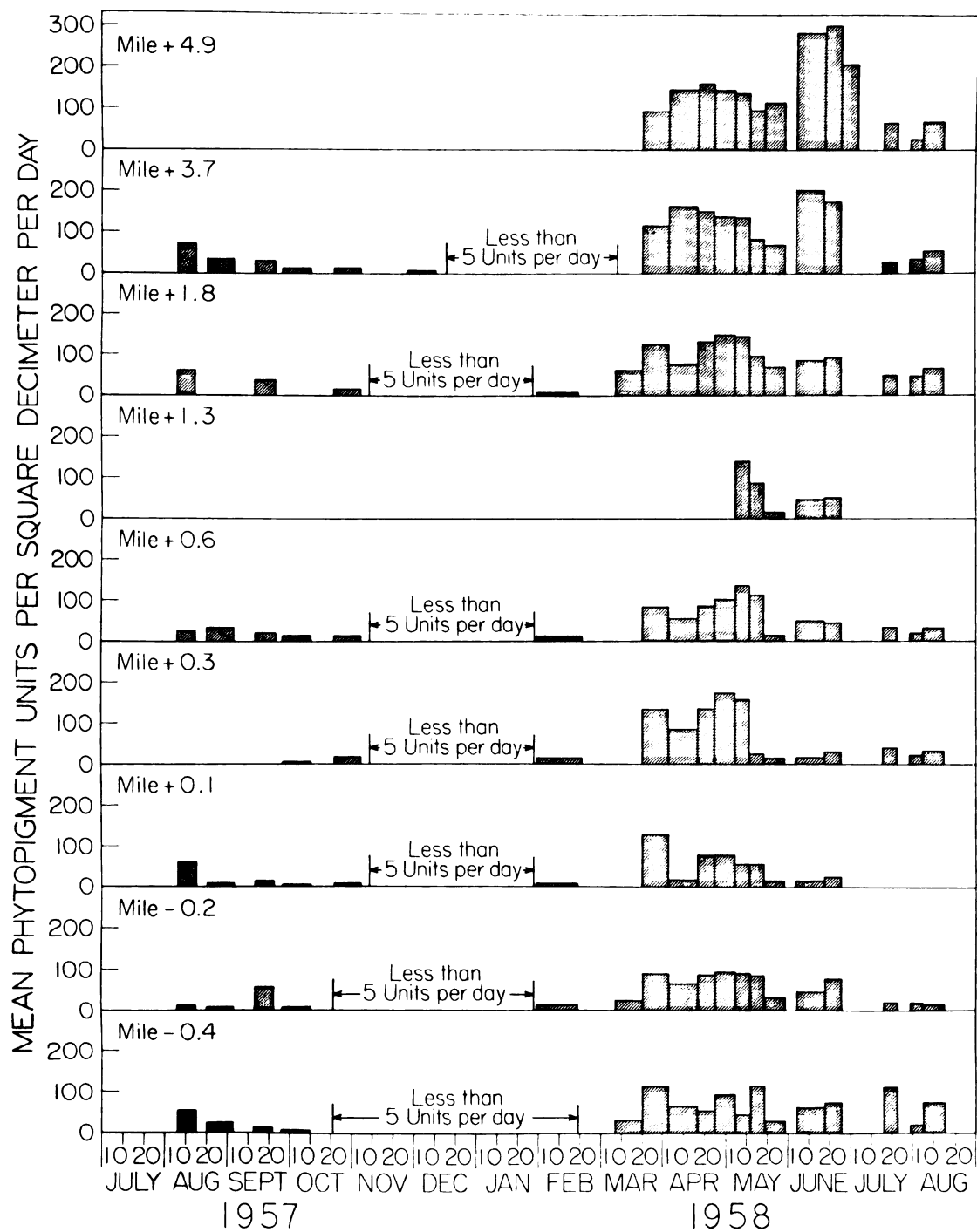
Qualitatively, the periphyton community of the Red Cedar River consisted primarily of members of the Bacillariaceae (diatoms). These organisms are characterized by their boxlike silica shells and yellow pigment which tends to mask the green chlorophyll. These diatoms become attached to the substratum by the whole of one surface of the cell or by means of a mucilaginous stalk or extrusion. Taxonomically, this is a very difficult group and since this study was undertaken to determine the relative production of organic material in the different areas, identification was not attempted.

Seasonal Variations

The periphyton production in the Red Cedar River demonstrated distinct seasonal variations (Fig. 15) which were difficult to correlate with the physical and chemical conditions of the stream.

The periphyton measurements were initiated August 6, 1957. The periphyton production during the first study period from August 6 to August 15, 1957 was relatively high with a mean phytopigment accretion of $78 \text{ AA} \times 10^3$ units per square decimeter per day at Station +3.7. The phytopigment production dropped approximately 50 percent during the next study period from August 20 to September 3, 1957 and continued to decrease until the period of ice cover in December. This indicates a decrease in the production of organic material through the fall period and the absence of the characteristic autumn maxima frequently recorded for a lotic environment.

Fig. 15. Mean daily phytopigment production per square
decimeter-measured at nine stations on the Red
Cedar River from August 1957 through August 1958.



During the winter period the ice cover on the Red Cedar River reached a maximum depth of 11 inches at Station +3.7 and was frequently covered with up to 6 inches of snow. This excluded a large portion of the available light from the organisms on the substrata and reduced primary production to the lower limits of detection possible by the phytopigment method of measurement. It should be pointed out that the depth of ice cover increased toward the downstream stations and this was reflected in a decrease in phytopigment production. The increase in the depth of the ice cover toward the downstream stations could not be correlated with differences in water temperature or stream flow.

The periphyton production increased very rapidly following the spring thaw and reached a maximum in April and early May. The phytopigment production at the downstream station exceeded 130 units per square decimeter per day during this period. A slight decrease in production was measured during the last half of May and then a sharp increase occurred during the month of June. A measurable quantity of ammonia nitrogen was found in the river at all stations during the June surge in production and, since it had been determined that during periods of high periphyton production the utilization rate of ammonia nitrogen was equal to or exceeded the production of ammonia, it was concluded that the source of this nutrient form was not indigenous to the stream. Although Resnick et al. (1958) found that cyanates were relatively stable under aerobic conditions, no other possible source of ammonia could be found. It is therefore assumed that the cyanates released into the river from the industrial plant lagoon at Fowlerville were carried downstream by the current and slowly underwent hydrolysis.

The ammonia produced by this reaction stimulated the periphyton growth. The greatest biological response was measured in the zone of phosphorus regeneration.

The periphyton production was receding as the extra available nitrogen supply was being utilized, when, for the second straight year, a heavy rainfall during the first week of July resulted in high water levels and high stream turbidities. As will be described later, the flood water conditions which existed for only a short period of time almost completely destroyed the standing crop of periphyton that was present immediately prior to the period of rainfall. After the turbid conditions had subsided, the periphyton production was comparable to the values measured during the late summer and early fall of 1957.

In summarizing the seasonal variations in periphyton production in the Red Cedar River, the data indicate that growth and reproduction increase rapidly following the period of ice cover and reach a maximum in April or early May. If the period when the extraneous nitrogen supply is ignored, it would appear that the production would slowly decrease through the summer and early fall until the period of ice cover when it is sharply reduced.

Variations between Stations

The phytopigment data indicate a discrete difference in the periphyton production between the stations located above and below the Williamston sewage treatment plant outfall, but the area of greatest biological response to the nutrients acquired from the sewage treatment plant effluent tends to shift with water temperature, stream velocity, and basic productivity levels (Fig. 15; Appendix C).

During the August to December period when the productivity was decreasing with each periphyton measurement period, the measured differences in production between stations became less pronounced. During the period from November 26 to December 10 the difference in organic material accumulation on the artificial substrata at the upstream and downstream stations showed very little difference. After the ice had formed on the river, no attempt was made to compare periphyton production and nutrient levels at the different stations. The data given in Appendix C illustrate the low level of growth during the winter months.

During the first periphyton collecting period after the ice period from March 7 to March 20, 1958, the artificial substrata were exposed only at Stations -0.4, -0.2, and +1.8 because of problems of accessibility. The mean phyt pigment accumulation for each station was 631, 445, and 1,222 units, respectively. The decrease in production at the second station was attributed to interference by turbid waters entering from Deer Creek whose confluence with the river is 0.1 mile upstream. This effect was noted during several periods throughout the course of the study and will be discussed in a later section.

The seasonal variations in periphyton production at the different stations are illustrated in Figure 16. In the spring when the water temperatures are relatively low, a significant response in periphyton growth was noted at the station 0.3 mile downstream from the outfall. Although the production level decreased towards Station +0.6, it frequently showed an increase further downstream. As the water temperatures increased, the area of greatest biological response to the acquired

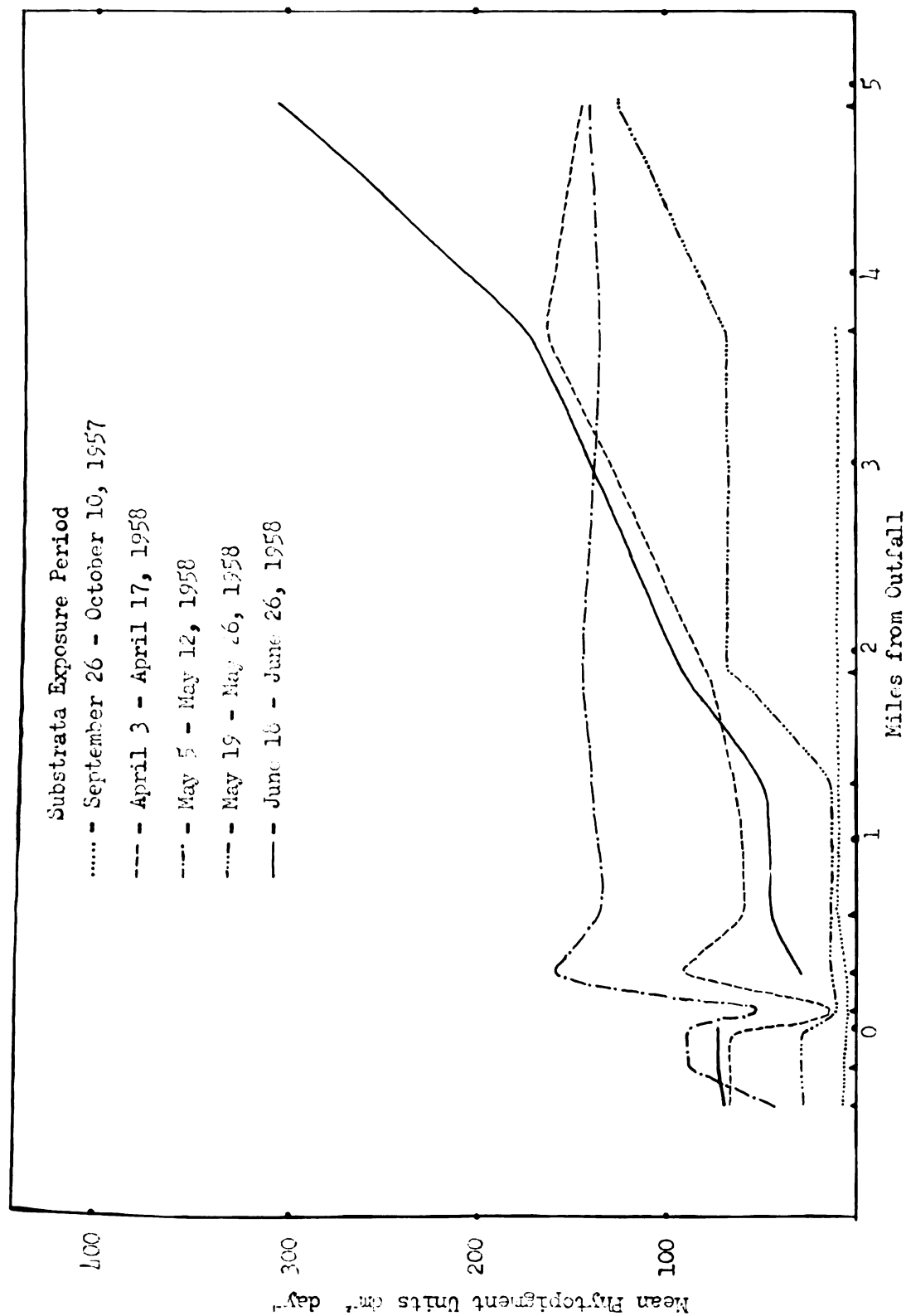


Fig. 16. Mean phytoplankton units per square decimeter per day for five collection periods from stations located above and below the sewage treatment plant outfall.

nutrients tended to shift downstream and it appeared that the sewage treatment plant effluent had an antagonistic effect upon the periphyton at Stations +0.1, +0.3, and +0.6. This is difficult to explain since no toxic industrial wastes are released into the river and heterotrophic organisms are not present in great enough numbers to compete for space with the periphyton on the substrata at the stations just downstream from the outfall. Also, Bartsch and Allen (1957) and others have reported that tremendous plankton populations are present in waste stabilization ponds where the medium is pure sewage in various stages of carbonization and nitrification. Although Brinely (1942) and Butcher (1947) reported a suppression of stream flora by sewage, the effects can be attributed to industrial wastes. Lackey (1956) found a sharp reduction in the plankton population of Lytle Creek after the introduction of sewage treatment plant effluent, but in this study the volume of effluent exceeded the stream flow and septic conditions resulted. At Station +0.3 of the Red Cedar River none of the above conditions exist, but the periphyton production showed a decrease during the summer months.

The relationship between the point of enrichment and the area of maximum biological response appears to be the result of interactions between several physical and chemical factors. The shift in the zone of highest periphyton production from Station +0.3 to the areas further downstream might be considered to be related to the water temperature. If this were true, the zone would move back upstream in the fall as the days became shorter and the water temperature decreased. This did not occur during the study period; in fact, the differences in the mean

phytopigment density measured at the different stations became less apparent as the fall progressed. This is demonstrated in Figure 16 when the mean water temperatures for the September 26-October 10, 1957 and the May 5-12 periods were approximately the same.

The decrease in periphyton production in the area between the sewage treatment plant outfall and Station +0.6 is not related to the phosphorus and nitrogen levels in the stream because the source is constant and the level of enrichment usually increases during the summer as the dilution factor decreases. Also, the nutrients in solution are utilized within the first 0.6 mile downstream from the outfall even during periods of low production. This indicates the periphyton community of the area is active and that the downstream response is not due to non-utilized nutrients originating from the sewage treatment plant outfall and flowing directly to the lower sections.

On the basis of the available data it is presumed that a substance or substances formed in the primary settling basin of the sewage treatment plant during the warm summer months inhibits plant reproduction in the stream down through Station +0.6. Although the periphyton growth and reproduction in this area is inhibited, the organisms present continue to remove the nutrient elements from the water and store them at levels far above their normal requirements. After death and detachment from the substratum, the cells are carried downstream by the current and undergo decomposition. The nutrient constituents are released to solution and produce an increase in periphyton production at the downstream stations. The nutrient supply during the early part of the year is also augmented by nitrates introduced with seepage and ground water.

During the fall period when the water temperatures are decreasing the antagonistic or inhibitory effect of the sewage treatment plant effluent on the periphyton population should also be reduced. This would allow a maximum periphyton growth in the areas just downstream from the outfall. A comparison of the spring and fall periphyton production periods having nearly equal water temperatures indicates that the fall production is much less than those measured in the spring even though the nutrient levels were comparable. A study of the "normal" solar radiation per square centimeter received by the area indicates that the fall values are only 75 percent of the spring values (computed from Grabb, 1950). Therefore, it may be assumed that even though other conditions may be equal, the flora of the stream cannot respond to the additional nutrients received from the sewage treatment plant under the lower light conditions.

The combination of the physical and chemical factors which produced the differences in periphyton production at the individual stations was the most obvious during June 1958 when the extraneous nitrogen supply became available to the flora of the study area.

The results of the chemical analyses of the water samples taken on May 27, 1958 indicated that the dissolved phosphorus concentration at Stations +1.8, +3.7, and +4.9 were 59, 68, and 71 $\mu\text{g l}^{-1}$, respectively. The value from Station -0.4 on this date was 23 $\mu\text{g l}^{-1}$. The total inorganic nitrogen content of the water at all the above stations was less than 0.1 mg l^{-1} .

The increase in available nitrogen during June produced a biological response that roughly paralleled the increase of regenerated dissolved phosphorus in solution toward the downstream stations (Fig. 15). During this period both nutrients were present in concentration above the minimal levels and the physical conditions were such that the periphyton could respond accordingly.

Nutrient response

It is usually difficult to detect limiting nutrient factors in a natural body of water and the Red Cedar River proved to be no exception. First, it is impossible to measure quantitatively only those nutrient compounds which are available to the plants. This is especially true in the case of a stream receiving nutrients in the form of sewage treatment plant effluent. The aquatic flora can utilize complex inorganic nutrient compounds as well as many organic forms. Chu (1946) reported that although pyrophosphate is not as good a phosphorus source as orthophosphate, it will promote growth. In the same study he found that the calcium or magnesium salt of inositol hexaphosphate (phytin) will support algae as well as orthophosphate. No data could be found concerning the status of the immediate availability to plants of the phosphorus in household detergents. In the routine method for phosphorus analysis either the orthophosphate only is measured or the sample is digested so that all of the phosphorus in phosphorus-bearing compounds is converted to the ortho- form for colorimetric detection. In a lake all of the forms of phosphorus are either immediately or potentially available to the flora but in a stream the potentially available forms should not be considered in a study area because they are carried downstream by the current before they are converted to an available form by biological or chemical action.

The aquatic flora can utilize also many forms of organic nitrogen which are not detected in the usual ammonia, nitrite, and nitrate-nitrogen analysis. Ludwig (1938) demonstrated that the green alga, Chlorella, can utilize organic nitrogen in the form of many of the

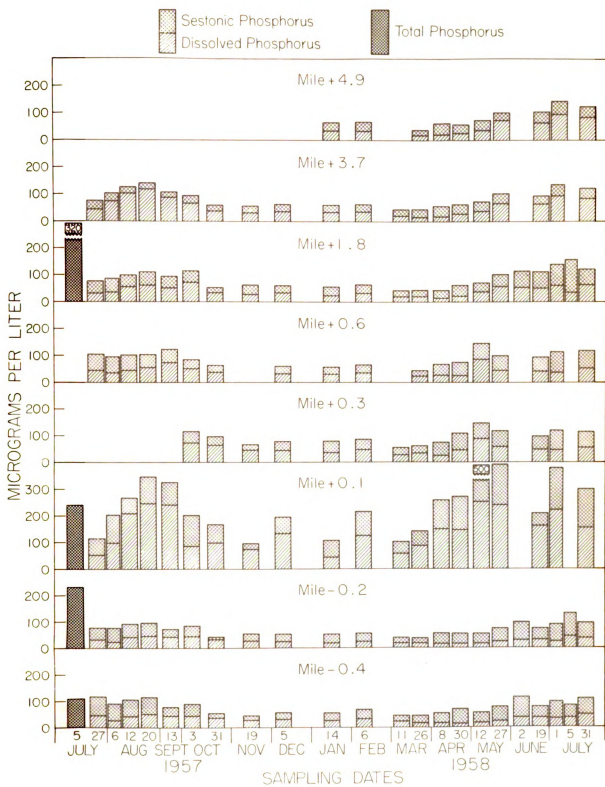
amino acids but cannot assimilate various azo compounds. Since it is known that the organic nitrogen content of sewage treatment plant effluent is very high and that nitrogen is a constituent of many complex compounds, the measurement of only that nitrogen which is immediately available to the flora of the stream is impossible.

Second, as pointed out by Whipple (1948), we are measuring only those nutrients which have not been used by the flora of the stream. The data of Kofoed (1903) indicate that the maximum plankton production in the Illinois River followed the highest nitrate values. In addition, it is well established that the inorganic nutrient content of a lake may approach the lower limits of detection at the time of a plankton bloom as a result of the available phosphorus and nitrogen being incorporated into living plants. Therefore, with a "cause and effect" phenomena controlling the nutrient levels, the chemical data must be interpreted very carefully before assuming that a particular element is a limiting factor in aquatic production.

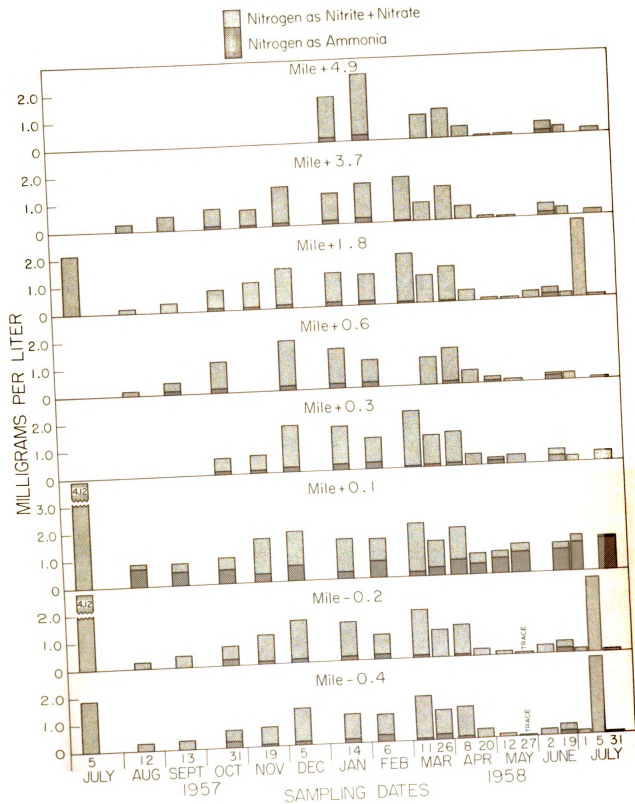
The chemical data indicate that the amount of soluble phosphorus present at all but two stations in the study area was less than the minimal levels for maximum algal growth (Chu, 1943) (Fig. 17). Assuming that antagonistic factors were present at Mile +0.1 and +0.3 during the summer period, one might conclude that phosphorus was the limiting factor for periphyton growth in the Red Cedar River. The production data for the entire study period tend to indicate that other factors are involved and although the soluble phosphorus levels are lower than the minimums given by Chu (op. cit.), they still exceed the concentrations in most unpolluted waters (Juday and Birge, 1931).

The inorganic nitrogen levels in the Red Cedar River for the July 1957 through July 1958 study period are given in Figure 18.

Fig. 17. Dissolved and sestonic phosphorus content of the
water at eight stations on the Red Cedar River.



**Fig. 18. Inorganic nitrogen content of the water at eight
stations on the Red Cedar River.**



The analyses of the periphyton production-nutrient level relationship for the individual study periods (Appendix C) indicate that available nitrogen may be of more importance as a limiting nutrient factor than the phosphorus. During the summer months when the water temperatures exceeded 20° C., the zone of phosphorus regeneration shifted upstream into the study area. This resulted in an increase in the dissolved phosphorus concentration in the water at Stations +3.7 and +4.9. Although the periphyton production usually responded to the higher nutrient levels, the increase in productivity was not sufficient to utilize the extra phosphorus that was available. Only during June 1958 when an extraneous nitrogen supply was introduced into the river did the periphyton produced at the downstream stations show a response adequate to utilize the regenerated phosphorus.

During several of the study periods an increase in periphyton production was accompanied by a decrease in the inorganic nitrogen content of the water. Furthermore, it was frequently noted that an increase in the available nitrogen supply was accompanied by a decrease in the dissolved phosphorus. The latter phenomenon was especially apparent at Station -0.2, downstream from the confluence with Deer Creek.

A comparison of the total phytopigment production and the mean nutrient levels for the 1958 portion of the study are given in Figure 19. The data indicate the necessity for considering all of the abiotic factors influencing the biodynamics of a stream.

Although the total phytopigment production more closely follows the phosphorus values than the nitrogen values, the rate of phosphorus uptake is not lineally related to the weight of periphyton produced.

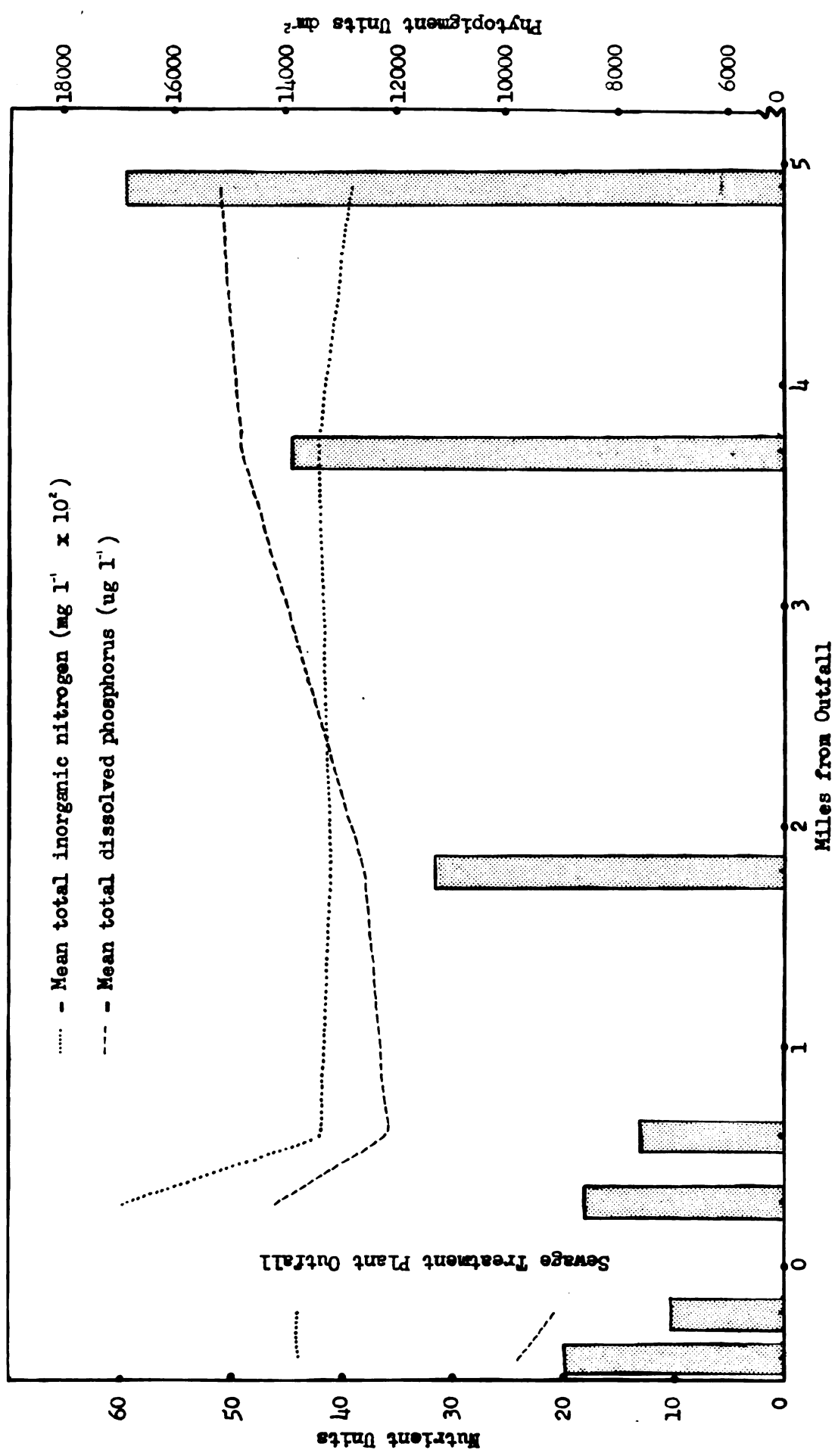


Fig. 19. Total accumulation of phytoplankton units per square decimeter of substratum and the mean available nutrient values recorded for the period from March 20, 1958 to August 15, 1958 at seven sampling stations on the Red Cedar River. Bars denote level of periphyton production.

The organic phosphorus:phytopigment ratio data given previously indicated that the flora in the areas immediately downstream from the sewage treatment plant outfall were removing more phosphorus per unit of organic matter than the flora either upstream from the outfall or in the downstream areas. Lund (1950) found that the phosphorus content of algal cells may vary by as much as a factor of 70. Harvey (1937) points out that large quantities of iron and phosphorus become adsorbed on the surface of diatoms. A portion of this adsorbed phosphorus is taken into the cell by the acidic protoplasm. Goldberg et al. (1950) found that as high as 50 percent of the phosphorus adsorbed on the surface of diatoms could be washed off with sea water and termed this phosphorus fraction as water-labile. Therefore, the quantity of phosphorus removed from the water by chem-adsorption and intracellular metabolism of periphyton might be very great in proportion to the amount of organic matter produced. Likewise, Gerloff and Skoog (1954) demonstrated that the nitrogen content of a blue-green alga could be more than doubled by increasing the nitrogen content of the media.

It follows then that a change in environmental conditions that retard growth might not necessarily result in an appreciable decrease in the rate of nutrient removal from the stream. Also, since the nutrient measurements involve only the excess elements that are not utilized by the aquatic flora, the element present in solution in the lowest relative concentration might be the most important as a limiting factor for periphyton production.

The decrease in phytopigment production from Station -0.4 to -0.2 is assumed to result from adverse physical conditions which frequently

occurred at the latter station due to the Deer Creek waters which enter the stream midway between the two areas. During periods of rainfall the tributary stream is quite turbid and, as will be discussed later, suspended solids proved to be very destructive to the standing crop of periphyton.

The decrease in mean phosphorus between the two areas may have been the result of the dilution effects by the tributary waters which were very low in phosphorus during normal flow periods.

Station +0.1 was not considered in Figure 19 because the effluent is not completely mixed with the river water in this area and the chemical data are not truly representative of the water to which the substrata were exposed. During the summer months there was also some interference by the sewage bacterium Sphaerotilus sp.

The periphyton at Station +0.3 did not respond to the increased available nutrients during the summer months. The rate of nutrient removal from the water apparently continued to remain high even though the periphyton growth was inhibited. This is indicated by the decrease in the quantity of the two essential elements between Stations +0.3 and +0.6. This may be explained in part by the previous discussion on nutrient storage and adsorption and the data on the organic phosphorus: phytopigment ratio at the different areas within the study section. The decrease in the phytopigment production at Station +0.6 parallels the decrease in the available nutrients of the station. Since both the phosphorus and nitrogen decrease so sharply, it is difficult to determine which might be termed the limiting factor. During two of the study periods, May 5-12 and May 12-19, the highest phytopigment production

was measured at this station. An examination of the chemical data from samples taken April 30, May 12, and May 19 indicate the dissolved phosphorus content of the water was abnormally high on May 12 ($84 \mu\text{g l}^{-1}$) but on the other sampling dates it was "normal" for the area. No differences were detected in the inorganic nitrogen concentration during this period.

After May 19, 1958, the production and available nutrient values were low at Station +0.6 and the greatest chemical changes and periphyton production were measured in the areas further downstream.

In the area downstream from Station +0.6 the phosphorus and phytoplankton values increase and the nitrogen values show a slight decrease. If the biological response at the downstream stations was entirely due to the increase in available phosphorus, it appears that the increase in production would be sufficiently great to prevent the increase in the quantity of dissolved phosphorus in solution. This would seem to indicate that the decrease in the mean inorganic nitrogen values towards Station +4.9 results from the increase in plant growth, but that the quantity of available nitrogen is not sufficient to support a periphyton population of sufficient magnitude to utilize the excess phosphorus.

The fate of the nitrogenous compounds in the decomposing, detached organic material from the upstream areas is unknown. The inorganic nitrogen values did not increase in proportion to the phosphorus regeneration. In a previous discussion the storage and adsorption of phosphorus by algae were mentioned as being responsible for the atypical organic phosphorus:organic matter ratio at Station +0.3. It is assumed that the quantity of nitrogenous material undergoing nitrification in the decomposing periphyton was not great enough to exceed the biological

uptake in the downstream areas and produce a measurable excess.

The maximum biological response in the downstream areas chiefly responsible for the increase in production (Figure 19) occurred during two periods when the available nitrogen was high. During one of the periods (April 1958) the nitrogen in the stream was being replenished by run-off water from the drainage area and in the other (June 1958) the available nitrogen was originating from an extraneous source. The available nitrogen-phytopigment relationship for the April 18-25 period is given in Figure 20. The total dissolved phosphorus values for April 22, 1958 only increased $10 \mu\text{g l}^{-1}$ between Stations +0.6 and +4.9.

In conclusion the data indicated a greater periphyton growth response to high nitrogen levels than to high phosphorus levels. During the summer months when a large amount of nitrogen is organically combined in terrestrial plants in the watershed area, the stream developed a nitrogen deficiency which inhibited periphyton growth and resulted in an increase in the dissolved phosphorus content of the water.

Effects of Turbidity on Stream Periphyton

Turbid water conditions were considered as a factor which limited periphyton production on the Red Cedar River. The two stations were established upstream from the sewage treatment outfall to measure the effects of turbidity introduced into the stream by the Deer Creek water on the standing crop and growth rate of periphyton. The area upstream from Deer Creek (Station -0.4) was subjected to adverse conditions only after relatively heavy rains whereas the area downstream from the confluence (Station -0.2) was subjected to adverse conditions following each rainfall of sufficient quantity to produce run-off. The seasonal

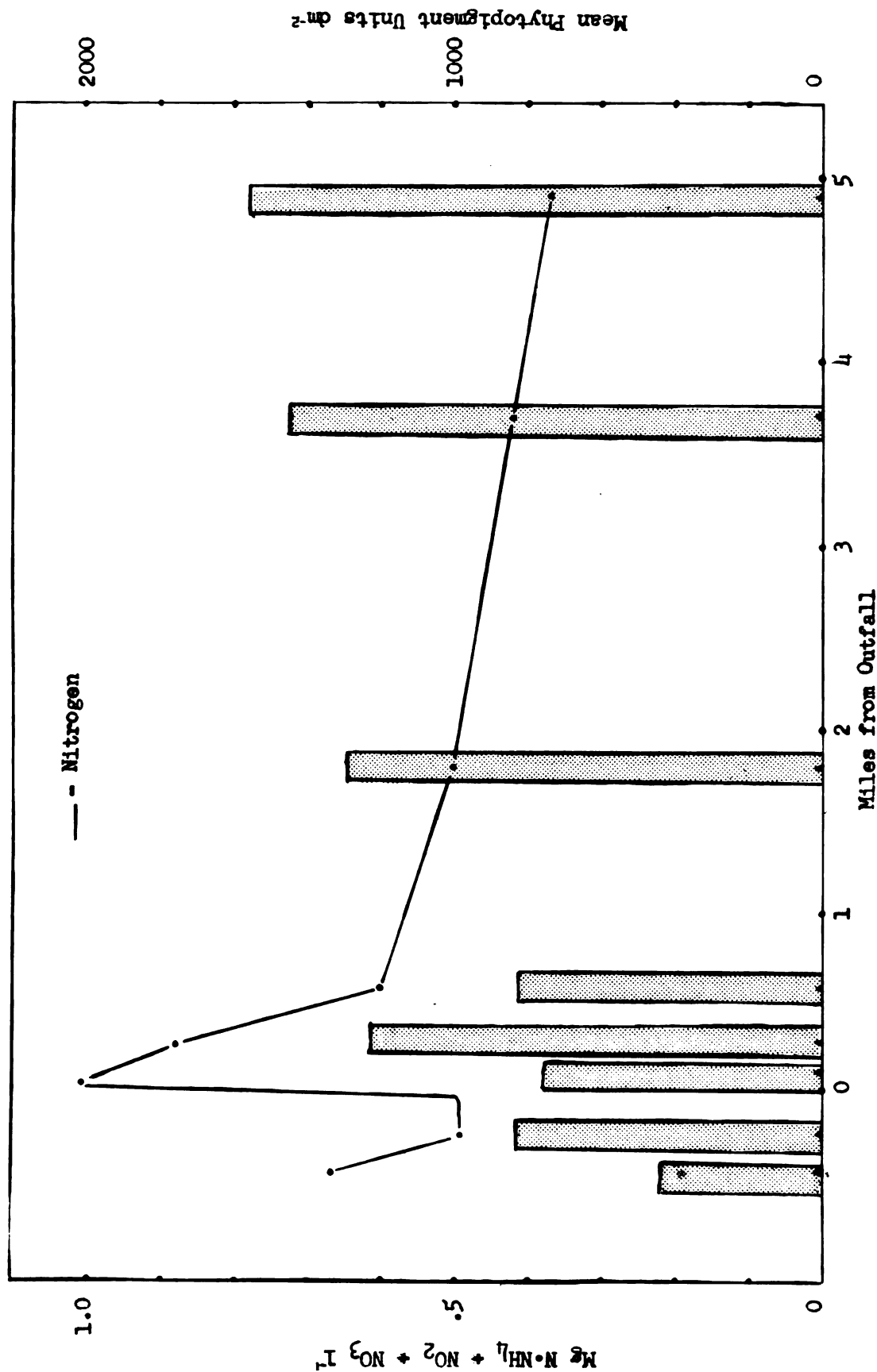


Fig. 20. Comparison of the inorganic nitrogen content of the water and the phytoplankton production measured at eight stations on the Red Cedar River for the period from April 18-25, 1958. Bars denote level of periphyton production.

reduction in periphyton production at the station downstream from Deer Creek has been discussed in a previous section and illustrated in Figure 19.

The heavy rainfall of July 3, 4, and 5, 1958 resulted in a four-foot increase in water level and turbidities which exceeded 150 units. Because of the extremely dry watershed, these adverse conditions existed on the stream for less than a week.

By chance the artificial substrata used to measure the periphyton production had been exposed for seven days prior to the rainfall period and were scheduled for removal on July 3. The substrata at Station +4.9 were removed from the stream on July 3 just prior to the rains and those from the remaining eight stations were allowed to remain in the stream for an additional seven days. This resulted in the units from Station -0.4 to +3.7 being exposed to stable water conditions for seven days which allowed for the establishment of a standing crop of periphyton on substrata. They were then exposed to approximately four days of severe adverse physical conditions which consisted of high stream velocity and high turbidity. This was followed by approximately three days when conditions were returning to "normal" (Fig. 14). This sequence of conditions provided data that clearly demonstrated the effects of turbid water conditions on the standing crop and growth of the periphyton within the study area of the Red Cedar River.

The measured phytopigment accumulation per square decimeter of substratum from the units removed after seven days exposure to stable water conditions at Station +4.9 were used to calculate the standing crop of periphyton present at the remaining eight stations prior to

high-water conditions. The production relationship between the stations was based upon the data obtained for the June 18-25 periphyton period. The calculated density for the entire 14-day period if normal stream conditions had persisted was estimated by multiplying the one-week densities by two. No information concerning the periphyton growth curve is available, but the values are thought to be within reason.

The substrata that were allowed to remain at the eight stations for the 14 days which encompassed the adverse physical conditions were removed from the stream on July 10, 1958. The phytopigments were extracted from the periphyton and the mean densities determined for each station. The measured values for the 7-day period for Station #4.9 and the measured values for the 14-day period from the remaining eight stations are plotted with the calculated values expected if stable water conditions had persisted in Figure 21.

The data indicate that the high water level conditions and associated turbidities which were present during July 4 to July 8, 1958 almost completely destroyed the standing crop of periphyton of the study area. The phytopigment density of the periphyton extract from the artificial substrata that were allowed to remain in the stream for the entire 14-day period can be considered to be from growth that took place between the time the turbidity had subsided and the substrata were removed from the river.

From this observation it is concluded that high-water levels and associated adverse conditions which may exist for only a short period of time can, through the abrasive action of the particles in suspension and exclusion of light, effectively destroy the standing crop of periphyton in a stream.

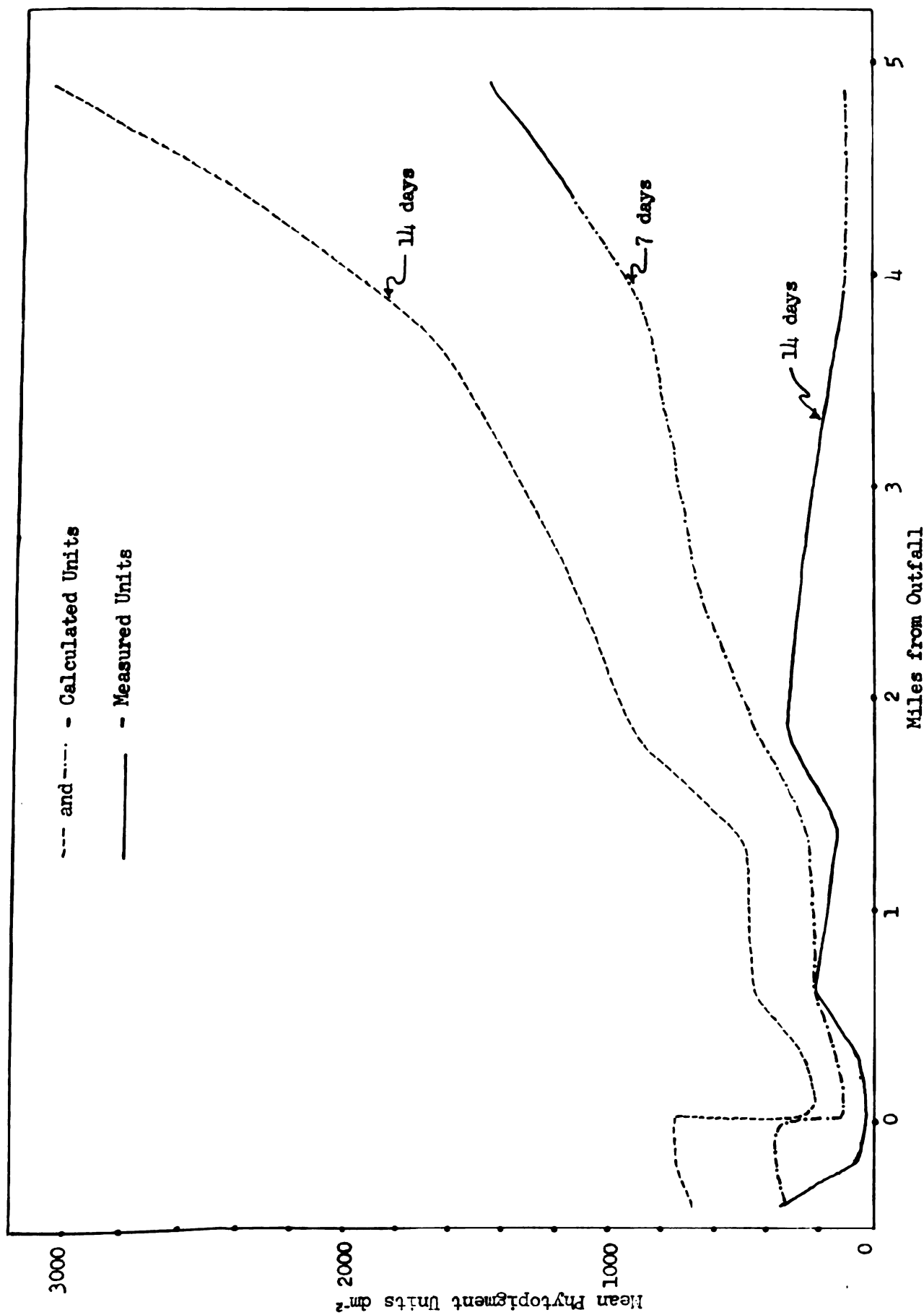


Fig. 21. Calculated and measured effects of high water and turbidity on primary production and on the standing crop of periphyton growing on plexiglass substrata in the Red Cedar River.

Spectrophotometric Analyses of the Phytopigments

The pigments extracted from periphyton with 95 percent ethanol are complex in nature and contain not only the chlorophyll group but also the xanthophylls, carotinoids, etc., and the alcohol soluble fats and oils. Having noticed a change in apparent color of the pigment complex from the different stations in the study area, it was theorized that the flora in the enriched areas might contain one of the minor pigments in sufficient quantities to produce a characteristic absorption spectrum and thus serve as a criterion for detecting organic enrichment or stream pollution.

A qualitative periphyton sample was collected from Stations -0.4 and +0.1 during the winter, spring and summer periods, the phytopigments extracted with 90 percent redistilled acetone, and the absorption spectra determined on a Beckman Model "B" spectrophotometer. The data indicate that a qualitative difference in the absorbancy spectra of phytopigment extracts from the different stations could not be detected until a growth of Sphaerotilus sp. was evident at Station +0.1. This sewage bacterium was first observed on July 3, 1958 and was thought to be associated with the high carbohydrate content of the sewage treatment plant effluent due to the home canning season. It was never observed at any of the other downstream stations during the study period.

The absorbancy peaks for a phytopigment extract complex from a periphyton community from the stream section unmodified by sewage treatment effluents are at 432 and 660 millimicrons. This is demonstrated in the spectrum of phytopigments from Station -0.4 (Fig. 22). The spectrum having absorbancy maxima at 410 and 665 millimicrons was

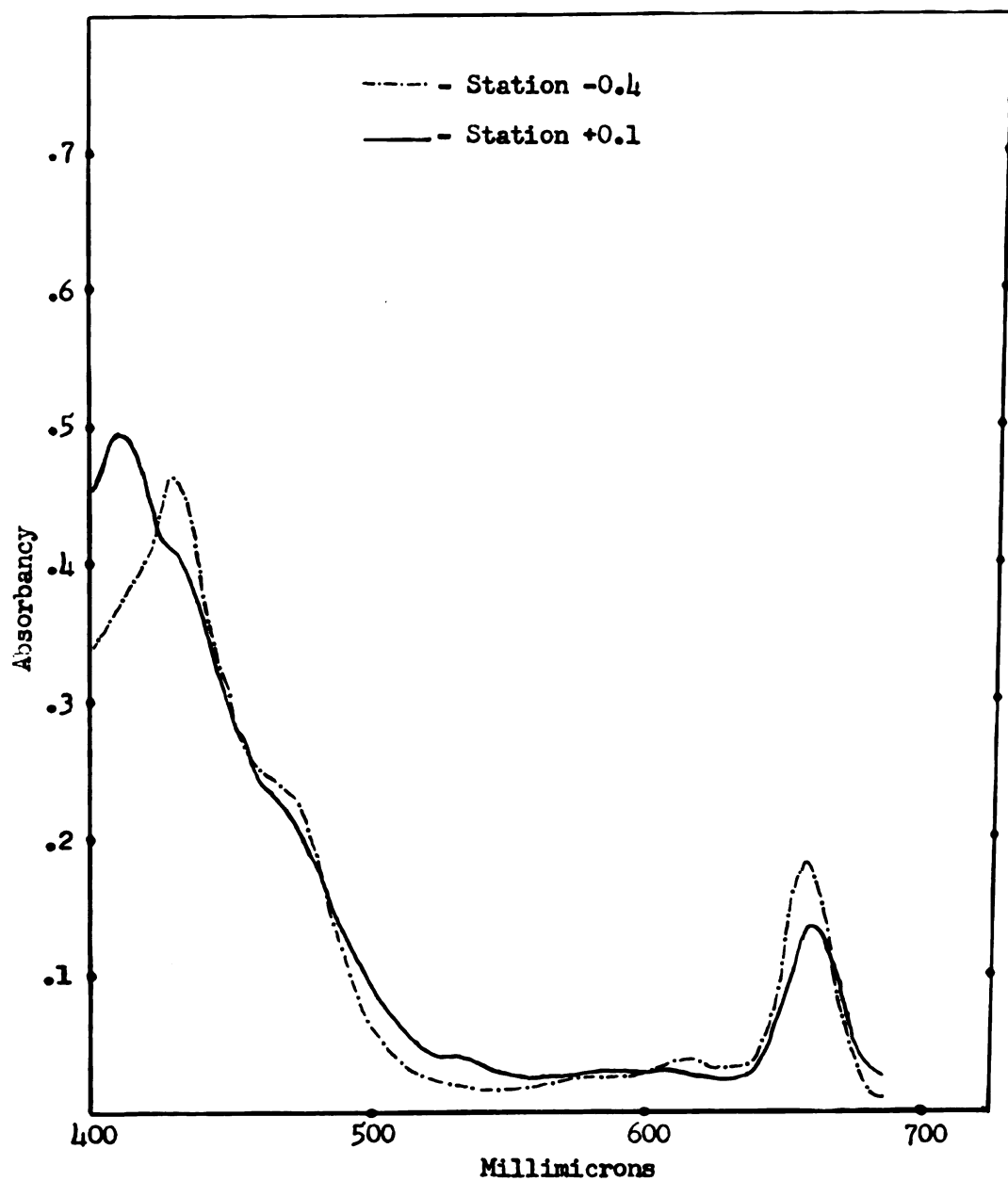


Fig. 22. Absorbancy spectra of the phytopigments from two stations on the Red Cedar River.

obtained from phytopigments extracted from periphyton collected at Station +0.1 on July 23, 1958. During this period a dense growth of heterotrophic organisms was noted on the artificial substrata in this area. The shift in the absorbancy maxima is attributed to the presence of decomposition products of chlorophyll (Holt and Jacobs, 1954). These products were formed after the heterotrophic growth destroyed the periphyton accumulation on the substrata.

In view of the above data it was concluded that a spectrophotometric analyses of phytopigment extracts could not be used to detect areas of organic enrichment or pollution in a stream until the population of heterotrophic organisms became sufficiently high to destroy the autotrophic community.

Projected Intercommunity Relationships

A complete study of the biodynamics of a lotic environment would involve the analyses of the physical and chemical factors which control the biological production plus a quantitative measurement of the biological production of each trophic level. Because of time limitations, the chemical analyses were confined to determining the phosphorus and nitrogen content of the water and the biological study involved only those organisms most closely dependent upon these nutrient elements.

Since Charles Elton first proposed the pyramid of numbers, the relationship between the higher biotic units and the primary producers has been studied and substantiated in both terrestrial and aquatic environments. Lindeman (1942) proposed the trophic-dynamic concept of ecology and stated that the caloric content of the organisms composing

each trophic level was homologous to the pyramid of numbers. Therefore, in view of the intensive and extensive work indicating a biological response of the higher organisms to increases in primary production, the scope of this investigation was limited to the basic producing units in the environment and the data used as an index to the production within the higher trophic levels.

The relatively short section of stream involved in the study above the sewage treatment plant outfall (0.4 mile) presented two distinct types of habitat. In the area below the Deer Creek confluence, the organisms were subjected to periodic, highly adverse conditions during high-water stages. The flood waters from Deer Creek arise in part from cultivated land and are highly turbid and carry a heavy bed load of sediments. The fine particles in suspension were detrimental to the flora and fauna in that they exclude light from the former, and the larger particles (chiefly fine grains of sand) mechanically destroy the biota by their abrasive action. The primary producers are able to recover rapidly after conditions return to normal but the primary consumer population would probably remain low until recolonization was affected by drift from upstream areas or ovaposition by adults which had emerged from other areas. Therefore, the reduction in the measured primary production at Station -0.2 might well be indicative of a reduced population of benthic forms, not only as the result of less available food, but also as the result of adverse physical conditions.

Although the flora at Station -0.4 was subjected to periods of turbid water conditions, most of the coarser silt particles and sand

settle out in the upstream reservoir. It has been demonstrated that the periphyton in this area recovered more rapidly than at other areas following high-water conditions and it can be assumed that the benthic forms here were subjected to less adverse conditions.

It would be impossible to project the periphyton data to the fish population in the stretch of river between the sewage treatment plant outfall and the Williamston Dam because of the short distance involved and the ability of fish to migrate from areas of less favorable environmental conditions to areas of more favorable conditions. In addition, the dam acts as a barrier to upstream migrants and undoubtedly served to concentrate those species attempting to move upstream in the Station -0.4 portion of the area.

The flora in the immediate area downstream from the sewage treatment plant outfall tends to store the nutrient elements in excess of their basic requirements and this produces an intermediate area where, during certain periods of the year, the measurable nutrients in solution are comparable to or less than the levels upstream from the outfall. This raises the question as to whether the production of organisms in the higher trophic levels would increase at Station +0.3 and decrease again at Station +0.6. In reviewing the data it can be seen that the zone of maximum periphyton response shifted from the Station +0.3 mile area in early spring to the downstream areas during May and June. Since the benthic forms could not migrate with the shift in maximum periphyton production, it is assumed that the population would be controlled by the minimal conditions in the habitat. This was confirmed in an earlier study (Brehmer, 1956) in which the benthic association in an

area downstream from a point of organic enrichment was composed of fewer taxonomic groups than at other areas in the stream even though recognized adverse conditions did not exist.

The increase in total periphyton production in the downstream areas is significant in many respects. Butcher (1947) noted a sharp decrease in sessile organisms immediately below the point of pollution but the pollutants included tar acids, gas liquors, and organic chemical wastes which would suppress the population. In his study the first population maximum was measured eight miles downstream from the sewage outfalls. On the Red Cedar River the maximum production was measured at Stations +3.7 or +4.9 only during 8 of the 24 study periods from August 6, 1957 to August 15, 1958. The point most important in considering the relationship of periphyton production to the higher biotic forms is that the production in the downstream part of the study area was always relatively high during the summer months when the activity of the poikilothermic herbivores is the greatest. There were no periods when the daily periphyton production dropped to almost zero as was noted in the areas within a mile of the outfall. Also, the maximum periphyton response was noted during April and May, a period of the year when the benthic forms are in their final instars prior to emergence and the grazing rate is high. The biological characteristics of the Station +4.9 area were reported in a previous study (Brehmer, op. cit.). The benthic association was not only more complex in that a larger number of taxonomic groups were represented but also more of the forms present were in the groups considered to be more suitable and available as fish food.

In view of the extensive literature relating benthic fauna to fish production (Ball and Hayne, 1952), it would appear valid to assume that the downstream areas are capable of supporting a greater fish population than either the area upstream from or immediately downstream from the sewage treatment plant outfall. Katz and Gaufin (1952) found that not only the total number of species and individuals of fish increased with distance from the sewage outfall, but that the fish failed to move upstream towards the outfall during the winter months when the dissolved oxygen content of the water became tolerable. The data from this study indicate that even though septic conditions did not exist at Stations +0.1, +0.3, and +0.6, the fish food organisms might be limited by periods of very low periphyton production.

Therefore, if the fish and bottom fauna production of an area are related to the minimal periods of periphyton production, a study of this type using artificial substrata and the phytopigment density of the accumulated periphyton as an index might well serve to classify a stream or an area thereof as to its recreational potential.

SUMMARY

SUMMARY

1. The mean dissolved phosphorus content of the Red Cedar River at Station -0.4 was found to be approximately $30 \mu\text{g l}^{-1}$ (range - 13 to 53). The mean inorganic nitrogen content of the water in this area was 0.7 mg l^{-1} (range - trace to 1.86). The quantity of nutrients in solution varies with stream flow, water temperature, and primary production levels.

2. The greatest influx of nutrients from the tributary streams occurs shortly after the start of a rainstorm and the concentration decreases rapidly during which time the activity of the aquatic flora is suppressed by adverse physical conditions. Thus the greatest proportion of these nutrient elements leached and eroded from the watershed are lost from the ecosystem without producing a biological response.

3. A major portion of the phosphorus and nitrogen available to the aquatic flora is introduced into the Red Cedar River from municipal drains and sewage treatment plant effluents. The phosphorus accrual from the Williamston sewage treatment plant effluent exceeds $100 \mu\text{g l}^{-1}$ during periods of normal stream flow. The annual phosphorus accrual from this source is approximately 2.5 metric tons.

4. The inorganic nitrogen accrual from the Williamston sewage treatment plant effluent exceeds 0.5 mg l^{-1} during periods of normal stream flow. The organic nitrogen content was not determined.

5. The accrued nutrients are removed from solution by biological uptake and/or chemical adsorption or precipitation within 0.6 mile downstream from the outfall. During periods of high stream temperatures and normal flow the biologically combined phosphorus is again released to solution within the 4.9 mile study area by the decomposition of periphyton which becomes detached from the area below the outfall. The accrued nitrogen did not reappear in solution in the inorganic form within the study area.

6. The ratio of organic phosphorus to phytopigment in the periphyton was more than four times greater at Stations +0.3 than at -0.2. The ratio decreased with distance from the outfall. This indicates that the aquatic flora in the enriched area is storing phosphorus in amounts over and above their normal requirements.

7. The data indicate that under normal conditions the periphyton production would increase rapidly after the period of ice cover until a maximum was reached about May 1. The rate of productivity would then decrease gradually until the winter ice period which sharply curtails growth. During the study period an extraneous available nitrogen source produced a second productivity peak during June. This growth pattern might be altered from year to year by changes in the distribution of the annual rainfall.

8. The biological response to the introduced nutrients was greatest during periods of high production and least during periods of low production. Variations in the depth of ice cover at the different areas made comparisons impossible during the winter period. The point of greatest biological response tended to shift downstream with increasing

water temperatures. This was also associated with an apparent antagonistic action of the effluent towards the periphyton at Stations +0.1 and +0.3. The measured net phytopigments for the five-month ice-free period of 1958 indicated that the production decreased immediately below the point of introduction of the effluent, increased at Station +0.3, decreased at Station +0.6, and then increased rapidly toward Station +4.9. The total production at Station +4.9 was more than double that measured at Station -0.2, the first station upstream from the outfall.

9. The increase in production at Station +4.9 was accompanied by an increase in the mean total dissolved phosphorus in solution and a decrease in the mean inorganic nitrogen in solution. This is interpreted as indicating that nitrogen is the nutrient that limits periphyton production in the Red Cedar River and results in the accumulation of dissolved phosphorus in the downstream areas. This interpretation was supported during the June production peak, when, as the extraneous nitrogen was made available to the flora, the quantity of regenerated phosphorus in solution decreased.

10. The data indicate that adverse physical conditions in the form of high stream flows and accompanying high turbidities, even though they may last for only a short period of time, can completely destroy the standing crop of periphyton in a river. The flora quickly becomes re-established as stream conditions return to normal.

LITERATURE CITED

LITERATURE CITED

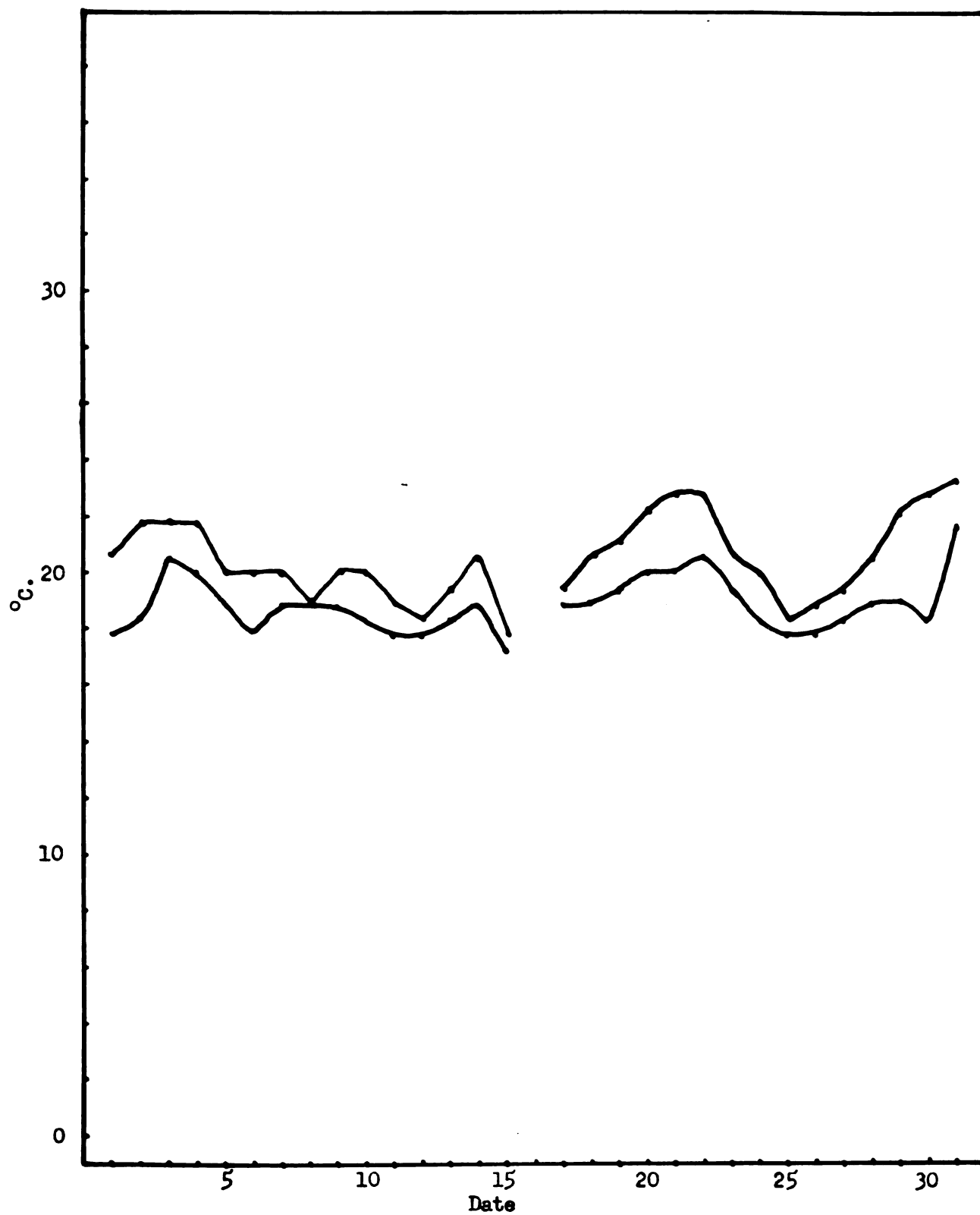
- Allen, K. R. 1951. The Horokiwi Stream. New Zealand Marine Dept. Fisheries Bull. No. 10, 231 pp.
- Alexander, G. R. 1956. The fertilization of a marl lake. (Unpub. Master's thesis, Michigan State Univ. Library).
- APHA, AWWA, FSIWA. 1955. Standard methods for the examination of water, sewage, and industrial wastes. 10th Ed. Baltimore, Waverly Press Inc. 522 pp.
- Ball, R. C., and D. W. Hayne. 1952. Effects of the removal of the fish population on the fish-food organisms of a lake. Ecology, 33: 41-48.
- Barrett, P. H. 1953. Relationships between alkalinity and adsorption and regeneration of added phosphorus in fertilized trout lakes. Trans. Amer. Fish. Soc., 82: 78-90.
- Bartsch, A. F., and M. O. Allum. 1957. Biological factors in treatment of raw sewage in artificial ponds. Limnol. and Oceanogr., 2: 77-84.
- Blum, J. L. 1957. An ecological study of the algae of the Saline River, Michigan. Hydrobiologia, 9: 361-408.
- Brinely, F. J. 1942. The effects of pollution upon the plankton population of the White River, Indiana. Invest. Ind. Lakes and Streams, 2: 137-143.
- Brehmer, M. L. 1956. A biological and chemical survey of the Red Cedar River in the vicinity of Williamston, Michigan. (Unpub. Master's Thesis, Michigan State Univ. Library).
- Butcher, R. W. 1932. Studies in the ecology of rivers. II. The microflora of rivers with special reference to the algae on the river bed. Ann. Bot., 46: 813-861.
- . 1947. Studies in the ecology of rivers. VII. The algae of organically enriched waters. J. Ec., 35: 186-191.
- Chu, S. P. 1943. The influence of the mineral composition of the medium on the growth of planktonic algae. Part II. The influence of the concentration of inorganic nitrogen and phosphate phosphorus. J. Ec., 31: 109-148.

- _____. 1946. The utilization of organic phosphorus by phytoplankton. J. Mar. Biol. Assoc. U. K., 26: 285-295.
- Cooke, W. B. 1956. Colonization of artificial bare areas by micro-organisms. Bot. Rev., 22: 613-638.
- Crabb, G. A. 1950. Solar radiation investigations in Michigan. Mich. State Agr. Exp. Sta. Tech. Bull. 222, 153 pp.
- Dobie, J., and J. Moyle. 1956. Methods used for investigating productivity of fish-rearing ponds in Minnesota. Minn. Dept. of Cons., Div. of Fish and Game. Spl. Pub. No. 5, 54 pp.
- Dobson, J. G. 1947. Disposal of cyanide wastes. Metal Finishing, 45: 78-81.
- Einsele, W. 1941. Die umsetzung von zugeführtem anorganischer Phosphat in eutrophen See und ihre Rückwirkung auf seinen Gesamthaushalt. Z. Fisch., 39: 407-488. (Cited from Biol. Abs., 23: 1046).
- Eldridge, E. F. 1933. Removal of cyanide from plating room wastes. Mich. State College Eng. Exp. Sta. Bull. 52, 20 pp.
- Ellis, M. M., B. A. Westfall, and M. D. Ellis. 1948. Determination of water quality. U. S. Dept. Inter., Fish and Wildlife Serv., Res. Rept. No. 9, 122 pp.
- Gerloff, G. C., and F. Skoog. 1954. Cell contents of nitrogen and phosphorus as a measure of their availability for growth of Microcystis aeruginosa. Ecology, 35: 348-353.
- Goldberg, E. G., T. J. Walker, and A. Whisenand. 1951. Phosphate utilization by diatoms. Biol. Bull. (Wood's Hole), 101: 274-284.
- Grzenda, A. R. 1955. The biological response of a trout stream to headwater fertilization. (Unpub. Master's thesis, Michigan State Univ. Library).
- Harvey, W. H. 1934. Measurement of phytoplankton population. J. Mar. Biol. Assoc. U. K., 19: 761-773.
- _____. 1937. The supply of iron to diatoms. J. Mar. Biol. Assoc. U. K., 22: 205-219.
- _____. 1940. Nitrogen and phosphorus required for the growth of phytoplankton. J. Mar. Biol. Assoc. U. K., 24: 115-123.
- Hentschel, E. 1916. Biologische Untersuchungen über den tierschen und pflanzenlichen Bewuchs in Hamburger Hafen. Mitt. Zool. Mus. Hamburg, 33: 1-172.

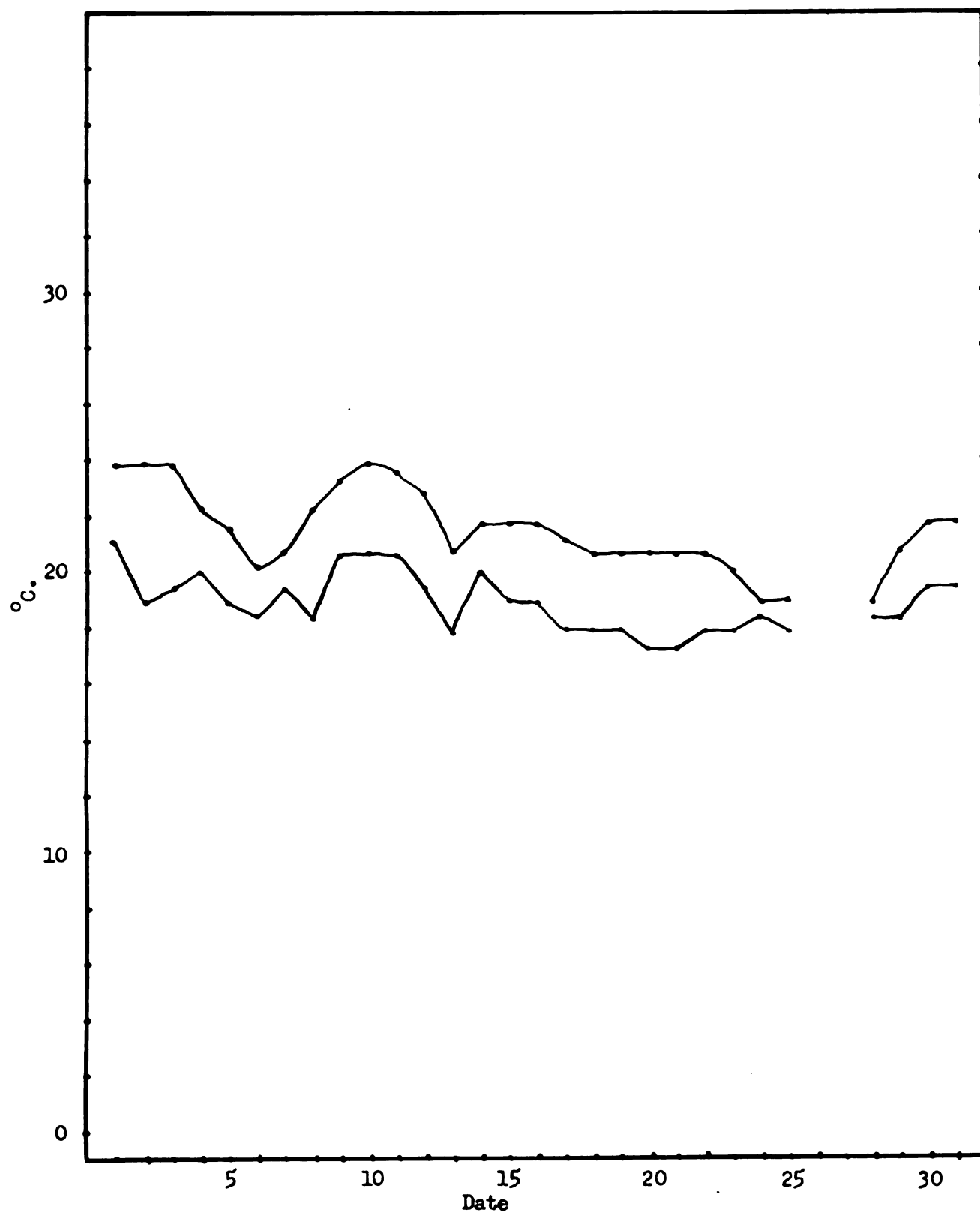
- Hepher, B. 1958. On the dynamics of phosphorus added to fishponds in Israel. *Limnol. and Oceanogr.*, 3: 84-100.
- Holt, A. S., and E. E. Jacobs. 1954. Spectroscopy of plant pigments. *Am. J. Bot.*, 41: 710-722.
- Hooper, F. F., R. C. Ball, and D. W. Hayne. (ms). Effects of fertilization of a marl lake.
- Huntsman, A. G. 1948. Fertility and fertilization of streams. *J. Fish Res. Board of Canada.*, 7: 248-253.
- Juday, C., and Birge, E. A. 1931. A second report on the phosphorus content of Wisconsin lake waters. *Trans. Wis. Acad. Sci. Arts Lett.*, 26: 353-382.
- Katz, M., and A. R. Gaufin. 1952. The effects of sewage pollution on the fish population of a midwestern stream. *Trans. Am. Fish. Soc.*, 82: 156-165.
- Kofoed, C. A. 1903. The plankton of the Illinois River 1894-1899, with introductory notes upon the hydrography of the Illinois River and its basins. Part 1. Quantitative investigations and general results. *Bull. Ill. State Lab. Nat. His.*, Vol. 6, Article II: 95-629.
- Kreps, E., and N. Verbinskaya. 1930. Seasonal changes in the Barents Sea. *J. de Conseil.*, 5: 327-345.
- Lackey, J. B. 1956. Stream enrichment and microbiota. *Pub. Health Rep.*, 71: 708-718.
- Lindeman, R. L. 1942. The trophic-dynamic aspect of ecology. *Ecology*, 23: 399-418.
- Ludwig, C. A. 1938. The availability of different forms of nitrogen to a green alga. *Am. J. Bot.*, 25: 448-458.
- Lund, J. W. G. 1950. Studies on Asterionella formosa, Hass.
II. Nutrient depletion and the spring maximum. *J. Ec.*, 38: 15-35.
- Manning, W. M., and R. E. Juday. 1941. The chlorophyll content and productivity of some lakes in northeastern Wisconsin. *Trans. Wis. Acad. Sci. Arts Lett.*, 33: 363-393.
- Moore, E. W. 1939. Graphic determination of carbon dioxide and the three forms of alkalinity. *J. Amer. Water Works Assoc.*, 31: 51-66.
- Needham, P. R., and R. L. Usinger. 1956. Variability in the macro-fauna of a single riffle in Prosser Creek, California, as indicated by the Surber sampler. *Hilgardia*, 24: 383-409.

- Patrich, R., M. H. Hohm, and J. H. Wallace. 1954. A new method for determining the pattern of the diatom flora. *Notulae Naturae*, 259: 1-12.
- Purdy, W. C. 1923. A study of the pollution and natural purification of the Ohio River. I. The plankton and related organisms. U. S. Pub. Health Bull., No. 13, 78 pp.
- Resnick, J. D., W. A. Moore, and M. B. Ettinger. 1958. Behavior of cyanates in polluted water. *Ind. and Eng. Chem.*, 50: 71-72.
- Richards, F. A., with T. G. Thompson. 1952. The estimation and characterization of plankton populations by pigment analyses. II. A spectrophotometric method for the estimation of plankton pigments. *J. Mar. Res.*, 11: 156-172.
- Rudolfs, W. 1947. Phosphates in sewage and sludge treatment. I. Quantities of phosphates. *Sewage Works J.*, 19: 43-50.
- Ruttner, F. 1952. *Fundamentals of limnology*. Univ. Toronto Press. 242 pp. Translated by D. G. Frey and F. E. J. Fry.
- Schmidt, E. L. 1956. Soil nitrification and nitrates in waters. *Pub. Health Rep.*, 71: 497-503.
- Taylor, D. M. 1937. The determination of phosphates in natural waters. *J. Am. Water Works Assoc.*, 29: 1983-1991.
- Whipple, G. S. 1948. *The microscopy of drinking water*. 4th Ed. New York, John Wiley and Sons. 585 pp.
- Whiteside, E. P., I. F. Schneider, and R. L. Cook. 1956. Soils of Michigan. *Mich. State Agr. Exp. Sta. Spec. Bull.* 402: 52 pp.

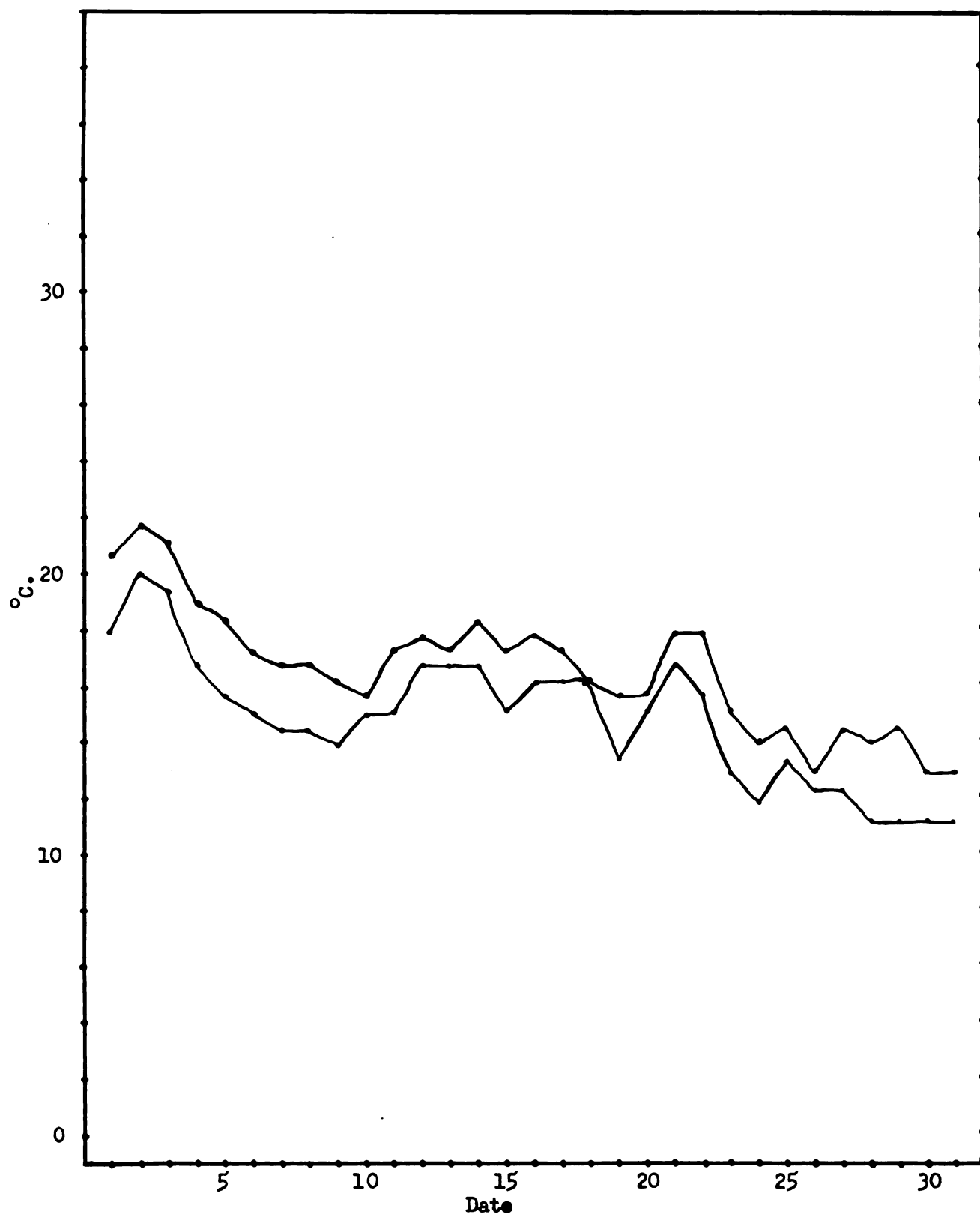
APPENDIX A



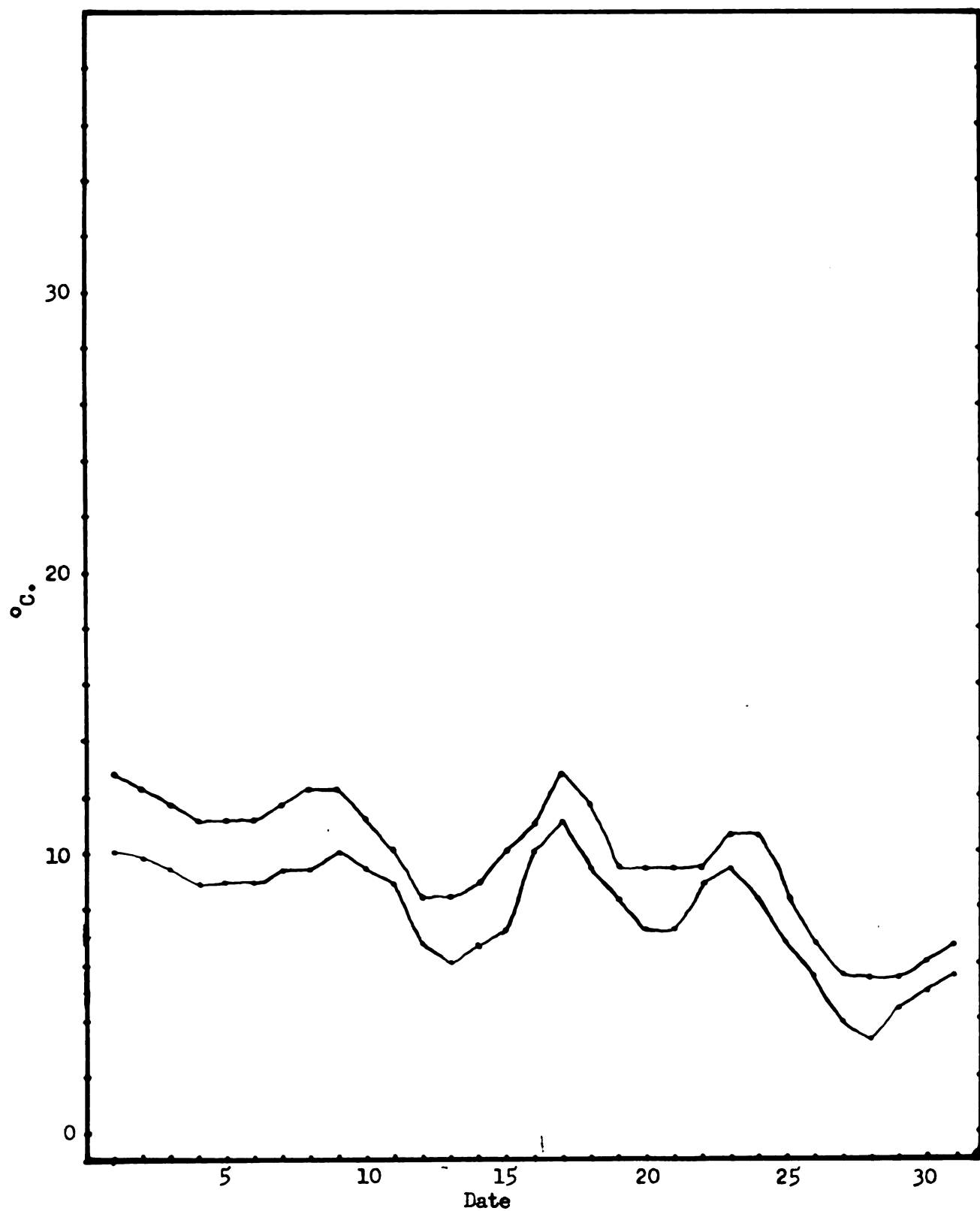
Daily high and low water temperatures recorded during JULY 1957 at a Station ten miles upstream from the mouth of the Red Cedar River.



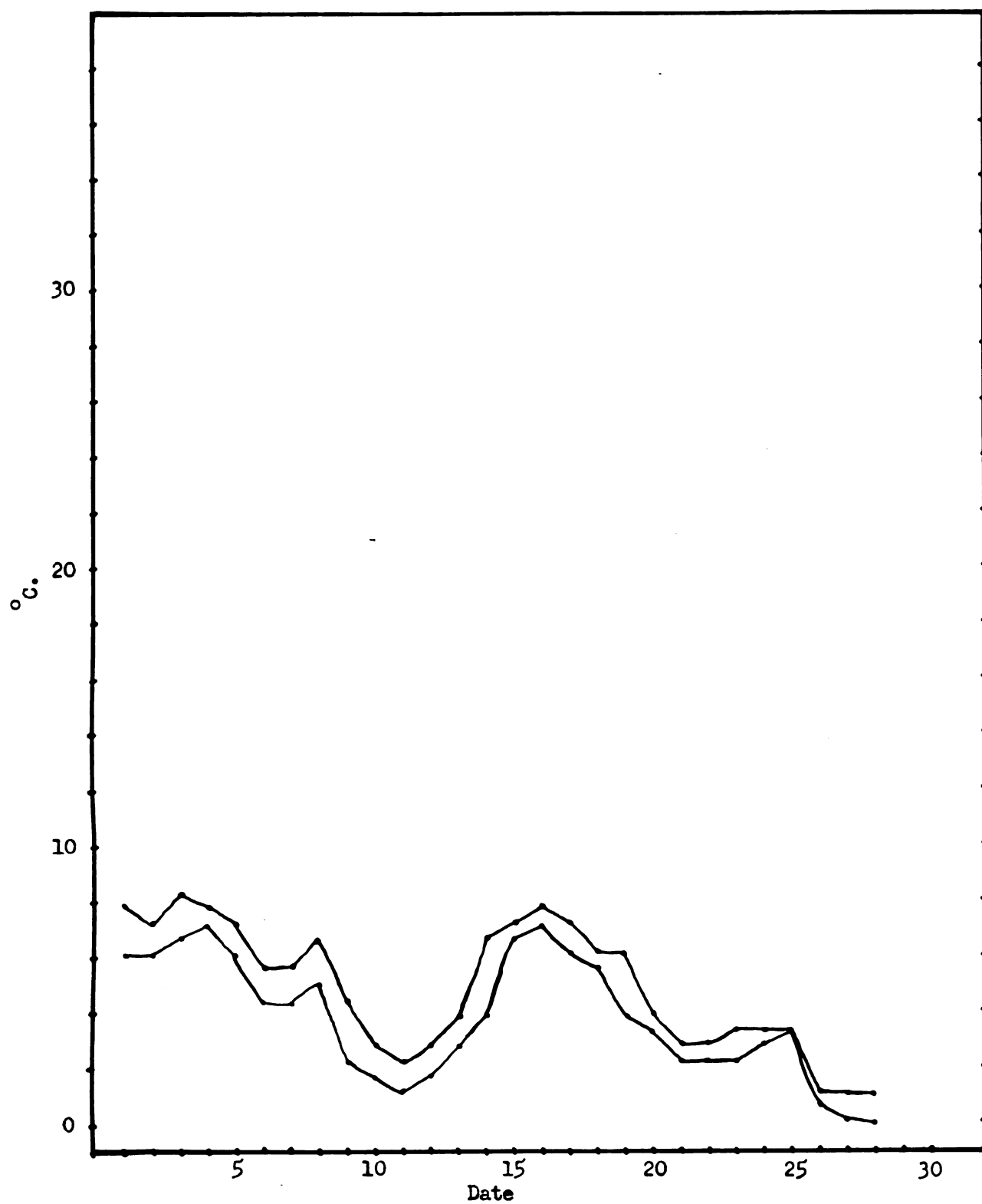
Daily high and low water temperatures recorded during AUGUST 1957 at a Station ten miles upstream from the mouth of the Red Cedar River.



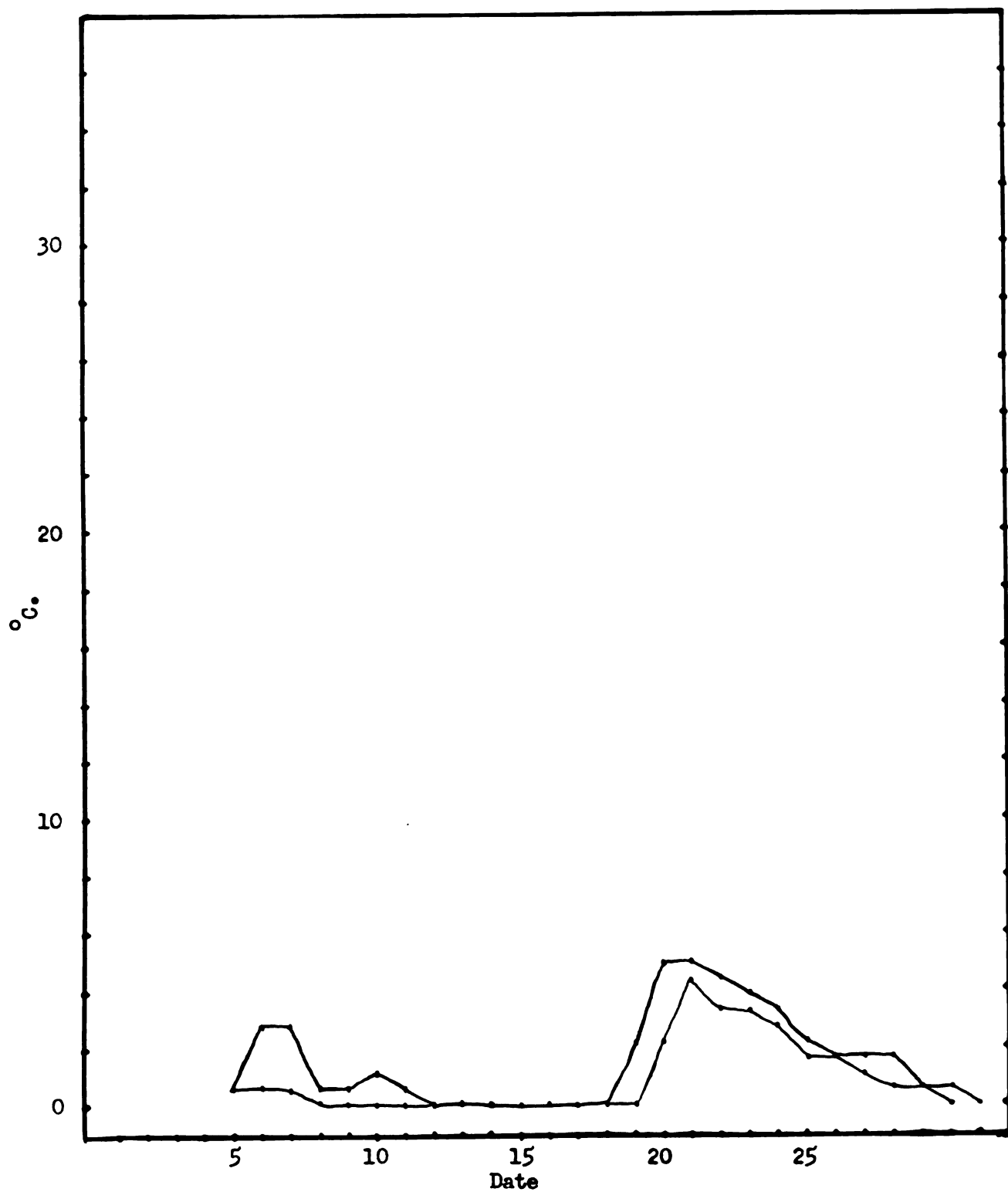
Daily high and low water temperatures recorded during SEPTEMBER 1957 at a Station ten miles upstream from the mouth of the Red Cedar River.



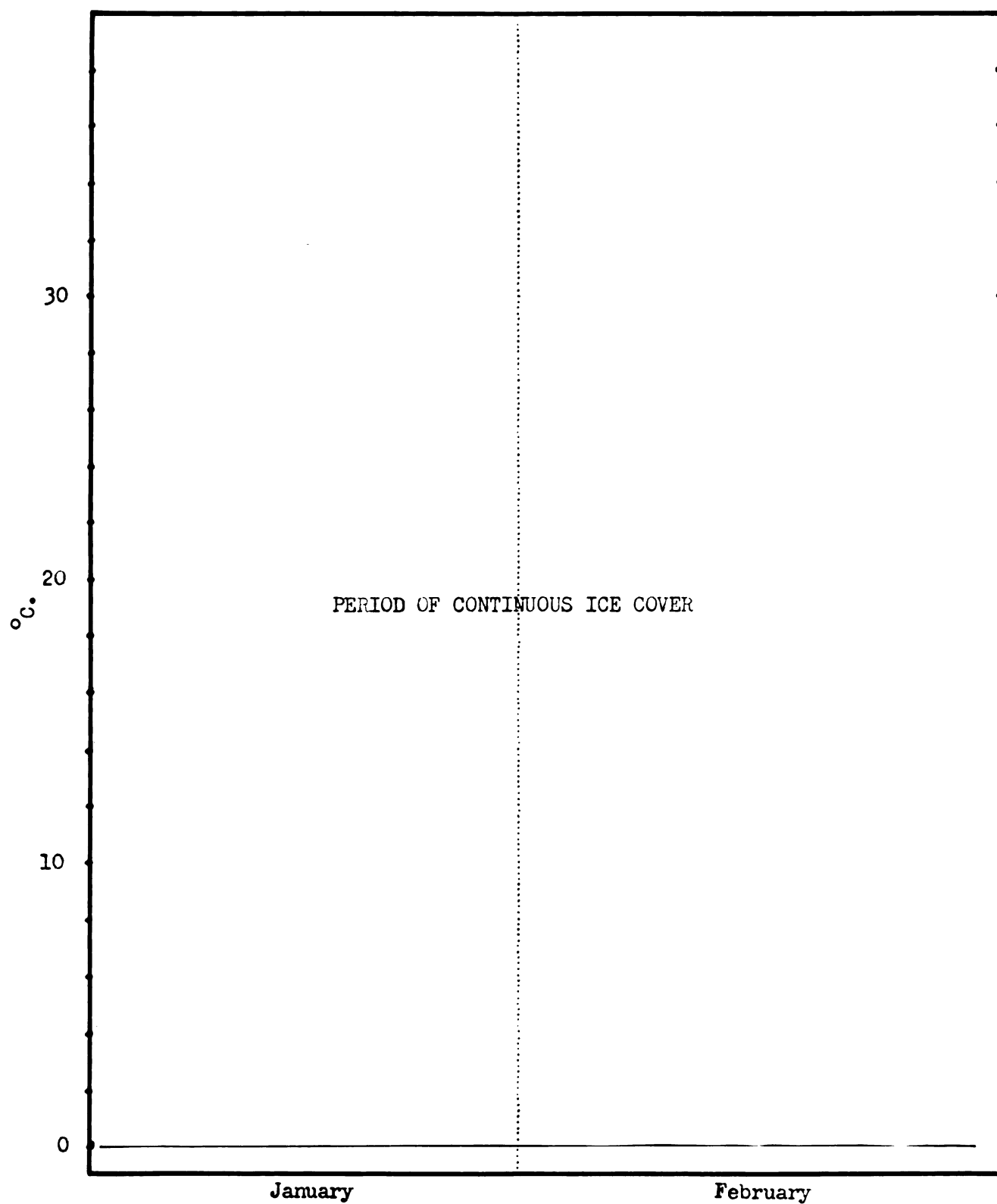
Daily high and low water temperatures recorded during OCTOBER 1957 at a Station ten miles upstream from the mouth of the Red Cedar River.



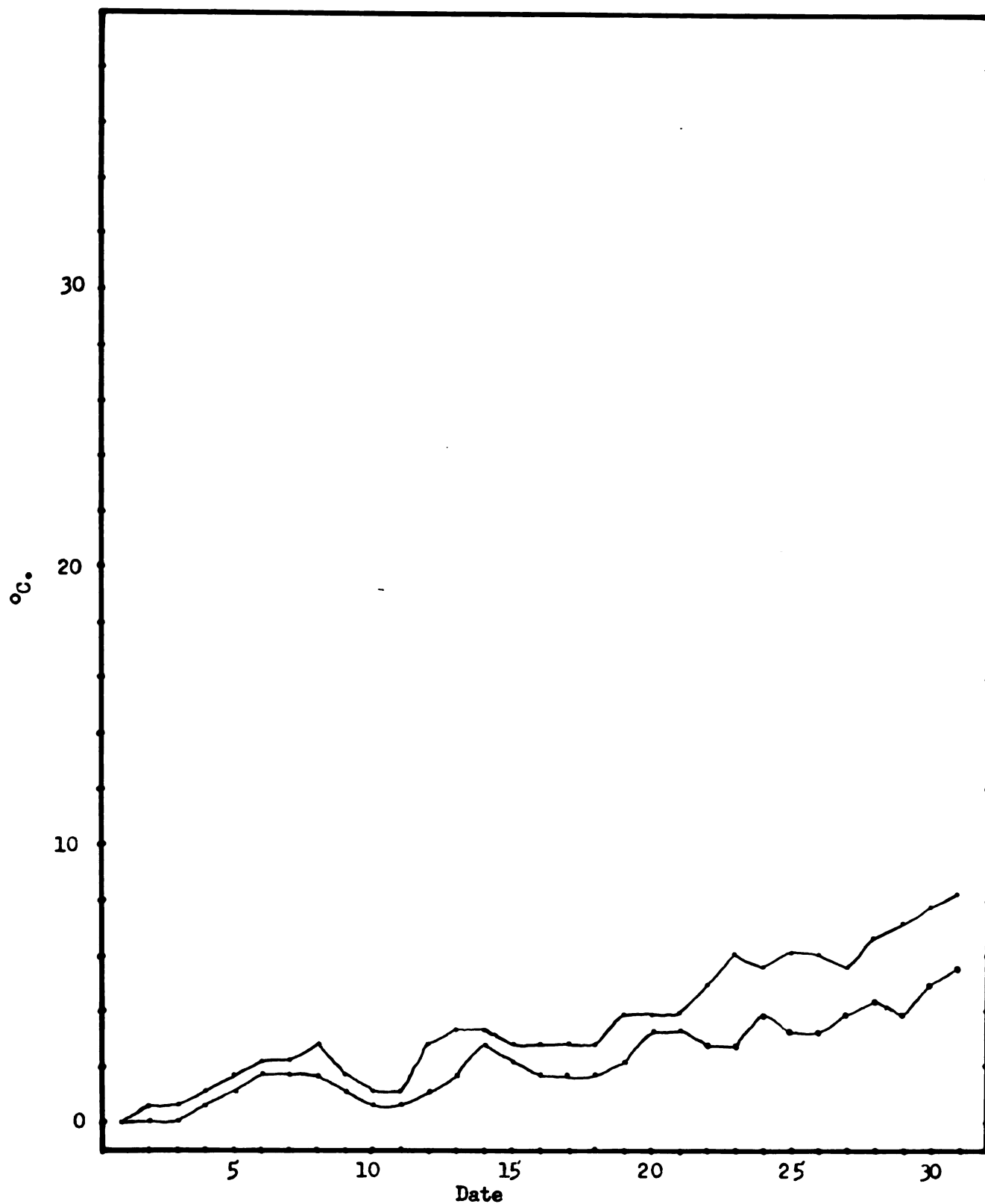
Daily high and low water temperatures recorded during NOVEMBER 1957 at a Station ten miles upstream from the mouth of the Red Cedar River.



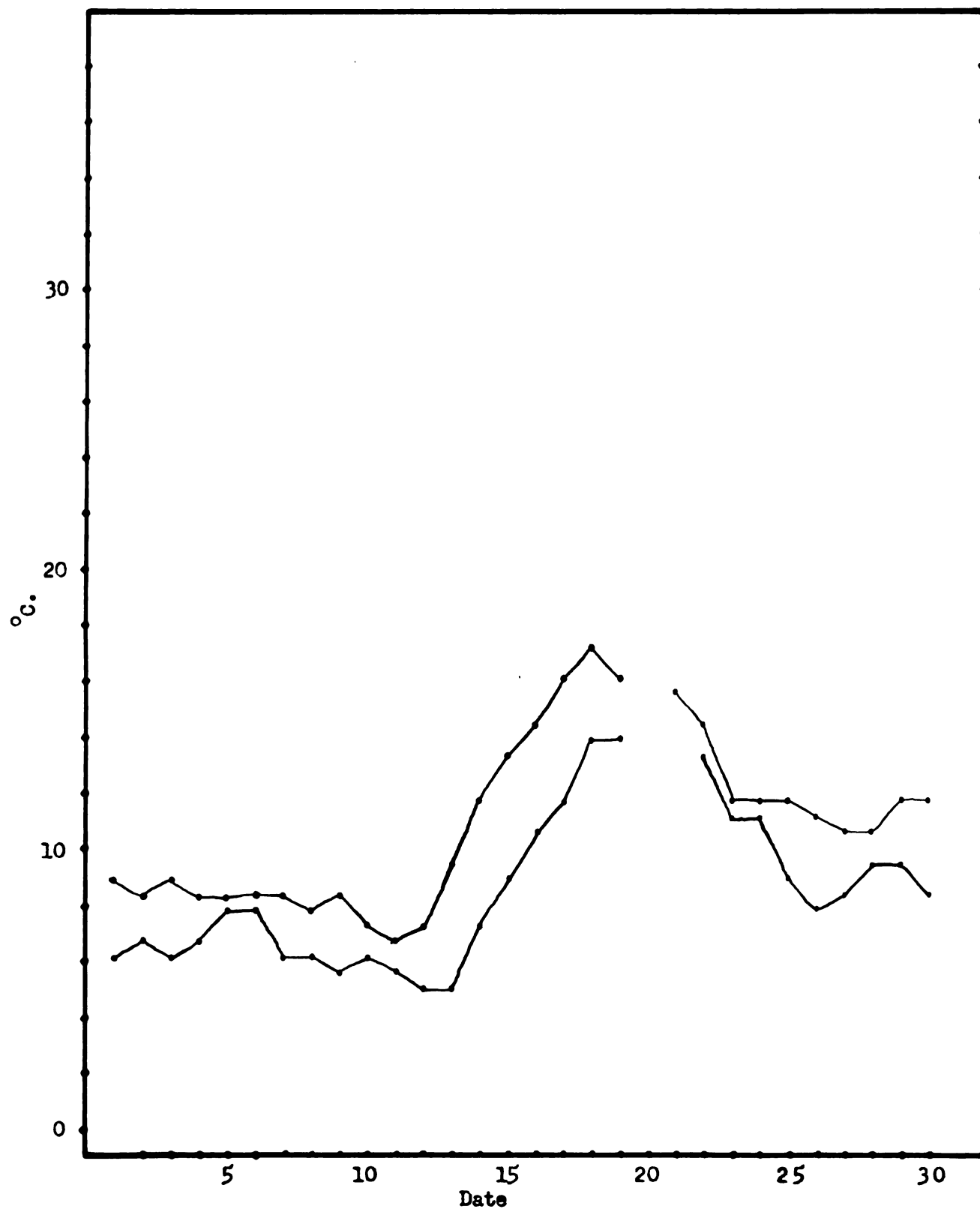
Daily high and low water temperatures recorded during DECEMBER 1957 at a Station ten miles upstream from the mouth of the Red Cedar River.



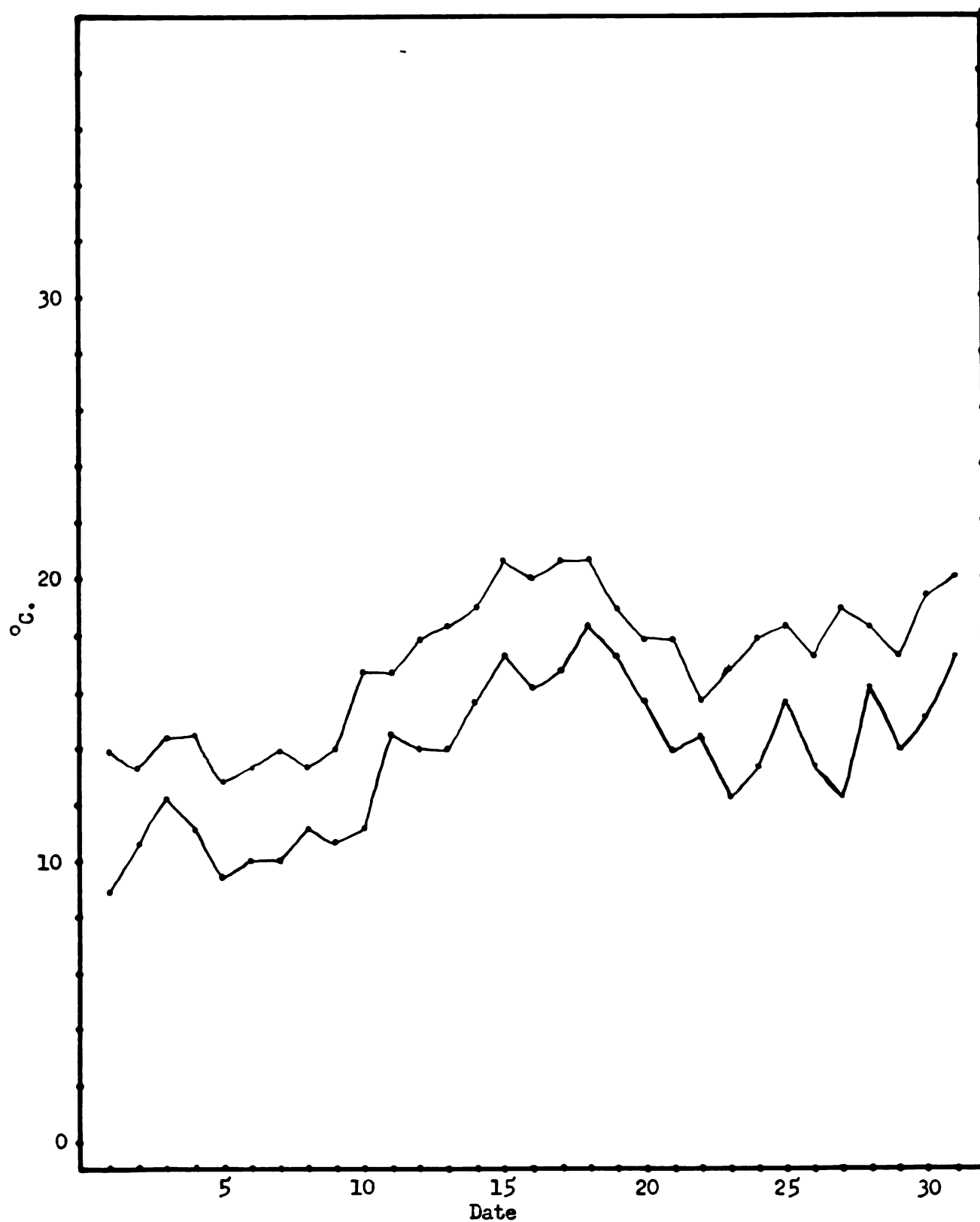
Daily high and low water temperatures recorded during JANUARY and FEBRUARY 1958 at a Station ten miles upstream from the mouth of the Red Cedar River.



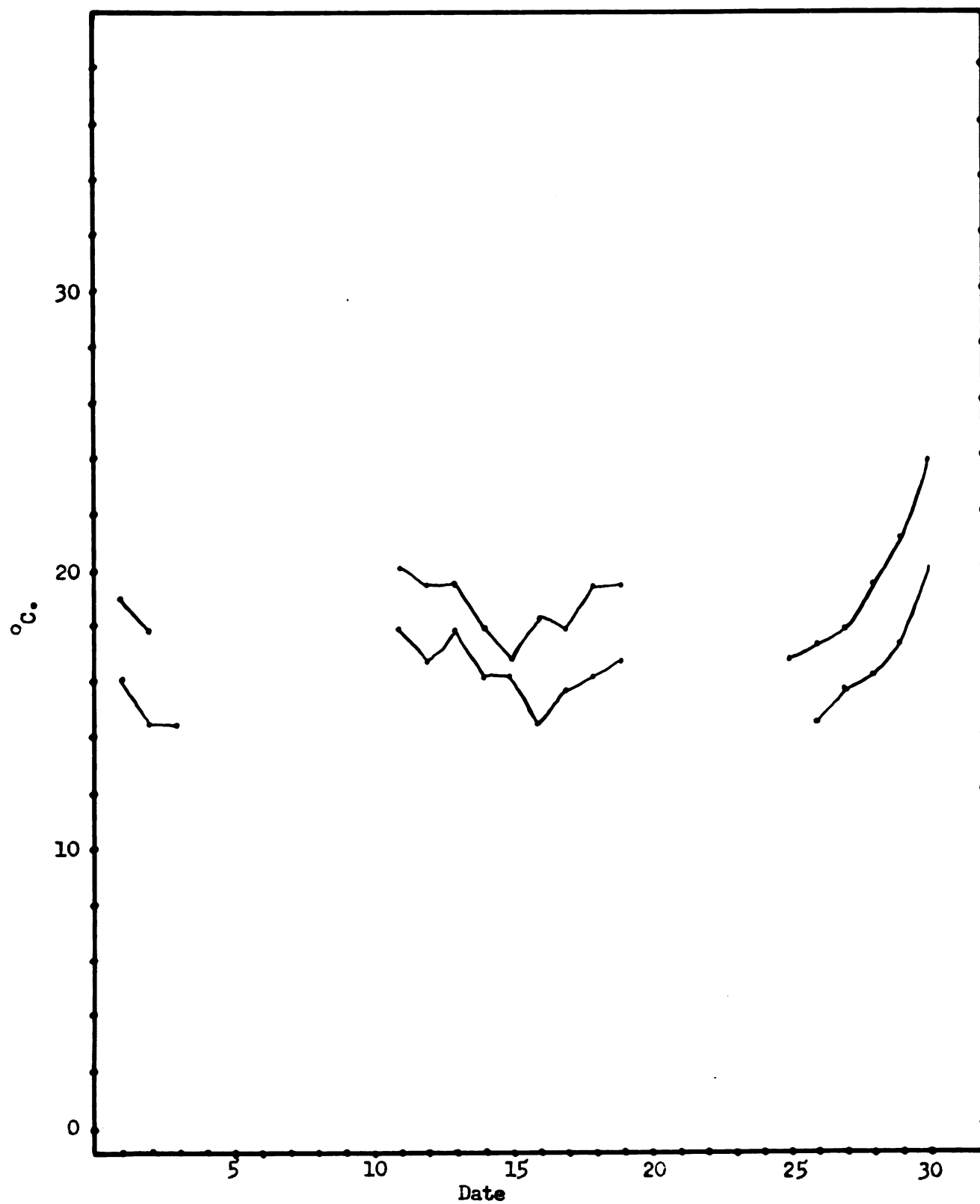
Daily high and low water temperatures recorded during MARCH 1958 at a Station ten miles upstream from the mouth of the Red Cedar River.



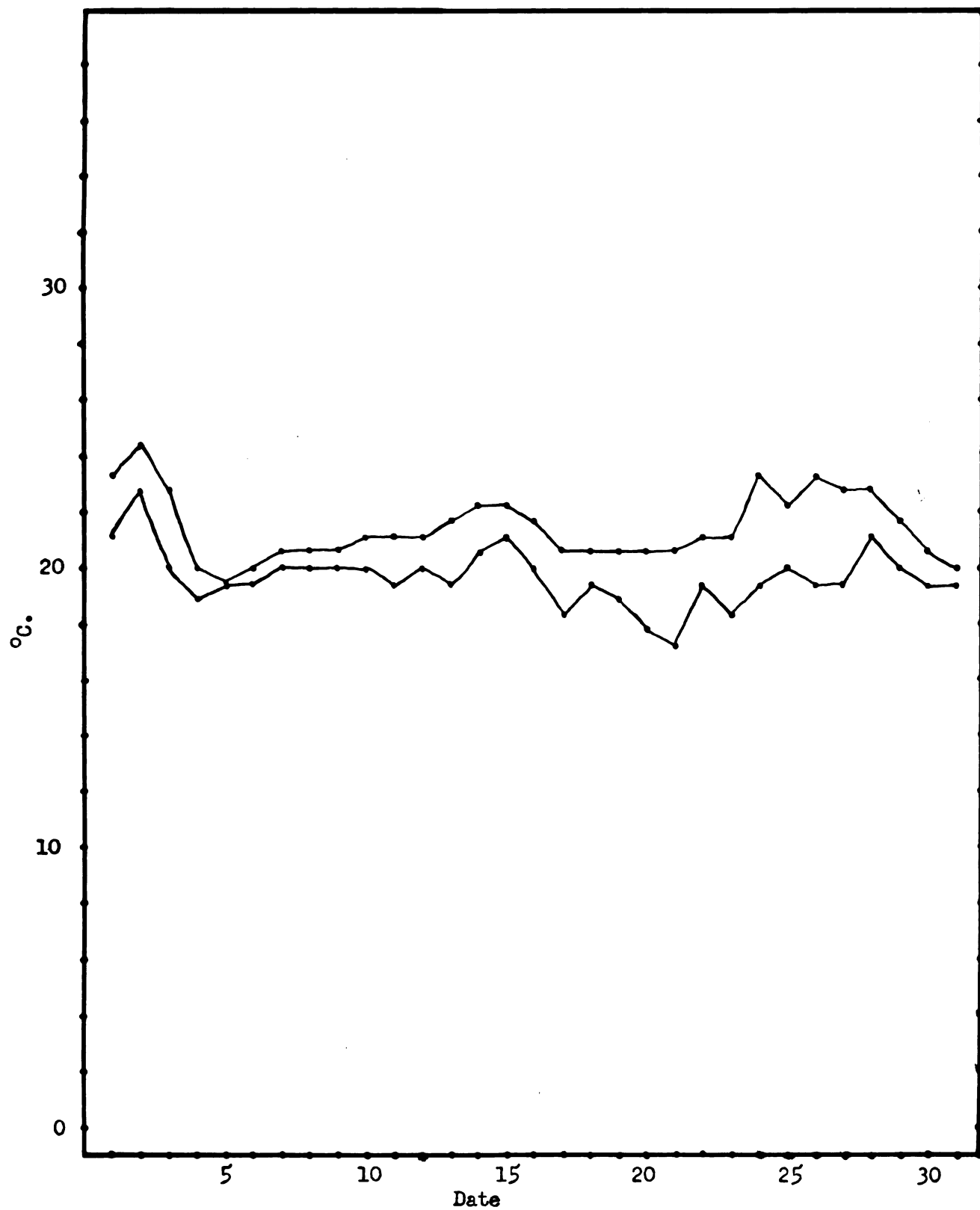
Daily high and low water temperatures recorded during APRIL 1958 at a Station ten miles upstream from the mouth of the Red Cedar River.



Daily high and low water temperatures recorded during MAY 1958 at a Station ten miles upstream from the mouth of the Red Cedar River.



Daily high and low water temperatures recorded during JUNE 1958 at a Station ten miles upstream from the mouth of the Red Cedar River.



Daily high and low water temperatures recorded during JULY 1958 at a Station ten miles upstream from the mouth of the Red Cedar River.

APPENDIX B

B-1

¹Data from U.S. Geological Survey, Lansing, Michigan Office

²Data from U.S. Weather Bureau, East Lansing, Michigan Office

Physical and chemical characteristics of the Red Cedar River - June 20, 1956

<u>Miles from mouth</u>	<u>4.5</u>	<u>7.8</u>	<u>10.3</u>	<u>13.3</u>	<u>16.6</u>	<u>18.3</u>	<u>18.8</u>	<u>23.5</u>	<u>27.1</u>	<u>31.3</u>	<u>34.3</u>	<u>34.7</u>
Gauge Height Mile 5 (feet)	3.83 ¹											
Discharge Mile 5 (c.f.s.)	202											
Air Temperature (°C.)	High - 30.4 Low - 17.1 ²											
Water Temperature (°C.)	23.0	22.5	22.5	22.0	21.3	21.5	21.5	21.0	20.5	21.5	21.0	19.0
Conductivity (x 10 ⁻⁶ ohms ⁻¹ cm ⁻¹)												
Alkalinity (mg l ⁻¹)	224	238	235	246	240	246	244	266	269	270	274	246
pH	8.0	8.1	8.0	7.9	7.9	7.9	7.9	7.8	7.8	7.8	7.8	7.9
Turbidity (units)	14	17	10	17	17	17	14	13	12	8	8	6
Dissolved Oxygen (mg l ⁻¹)	6.0	5.6	6.0	5.4	6.2	5.8	5.4	5.4	5.0	4.8	6.2	6.0
Total Phosphorus (µg l ⁻¹)	105	85	77	80	75	83	142	93	74	69	35	19
Nitrogen·NH ₄ (mg l ⁻¹)												
Nitrogen·NO ₂ + NO ₃ (mg l ⁻¹)												

¹Data from U. S. Geological Survey, Lansing, Michigan Office²Data from U. S. Weather Bureau, East Lansing, Michigan Office

Physical and chemical characteristics of the Red Cedar River - June 25, 1956

Miles from mouth 4.5 7.8 10.3 13.6 16.6 18.3 18.8 23.5 27.1 31.3 34.3 34.7

Gauge Height Mile 5 (feet) 3.64¹

Discharge Mile 5 (c.f.s.) 143

Air Temperature (°C.) High - 23.3 Low - 13.2²

Water Temperature (°C.) 22.5 21.5 21.5 21.5 22.0 22.0 22.5 19.5 18.5 18.5 17.5 17.0

Conductivity
(x 10⁻⁶ ohms⁻¹ cm⁻¹)

Alkalinity (mg l⁻¹) 252 254 258 262 266 264 260 260 268 268 266 262

pH 7.9 7.8 7.9 7.9 7.9 7.9 7.9 7.8 7.8 7.8 7.9 7.8

Turbidity (units) 13 18 9 8 8 13 15 15 8 9 5 6

Dissolved Oxygen (mg l⁻¹) 5.6 5.6 5.8 5.0 5.2 5.0 4.4 5.2 4.9 4.4 5.6 5.6

Total Phosphorus (µg l⁻¹) 165 31 61 21 41 96 240 94 59 91 48 26

Nitrogen·NH₄ (mg l⁻¹)

Nitrogen·NO₂ + NO₃ (mg l⁻¹)

¹Data from U. S. Geological Survey, Lansing, Michigan Office

²Data from U. S. Weather Bureau, East Lansing, Michigan Office

Physical and chemical characteristics of the Red Cedar River - June 30, 1956

<u>Miles from Mouth</u>	<u>4.5</u>	<u>7.8</u>	<u>10.3</u>	<u>13.6</u>	<u>16.6</u>	<u>18.3</u>	<u>18.8</u>	<u>23.5</u>	<u>27.1</u>	<u>31.3</u>	<u>34.3</u>	<u>34.7</u>
Gauge Height Mile 5 (feet)	3.46 ¹											
Discharge Mile 5 (c.f.s.)	93											
Air Temperature (°C.)	High - 30.4 Low - 15.6 ²											
Water Temperature (°C.)	24.5	24.0	25.0	--	--	21.0	21.0	20.0	20.0	19.0	18.5	18.0
Conductivity (x 10 ⁻⁴ ohms ⁻¹ cm ⁻¹)												
Alkalinity (mg l ⁻¹)	254	260	264	258	264	269	269	272	266	260	260	248
pH	8.1	8.2	8.2	8.1	8.1	8.0	8.0	8.1	8.0	8.1	8.1	8.0
Turbidity (units)	8	7	5	5	6	8	9	8	8	3	3	2
Dissolved Oxygen (mg l ⁻¹)	7.8	7.0	8.0	6.4	7.2	6.4	5.8	6.6	6.2	7.0	6.6	8.0
Total Phosphorus (µg l ⁻¹)	65	19	21	24	22	71	149	45	58	19	10	10
Nitrogen•NH ₄ (mg l ⁻¹)												
Nitrogen•NO ₂ + NO ₃ (mg l ⁻¹)												

¹Data from U. S. Geological Survey, Lansing, Michigan Office

²Data from U. S. Weather Bureau, East Lansing, Michigan Office

Physical and chemical characteristics of the Red Cedar River - July 5, 1956

<u>Miles from Mouth</u>	<u>4.5</u>	<u>7.8</u>	<u>10.3</u>	<u>13.6</u>	<u>16.6</u>	<u>18.3</u>	<u>18.8</u>	<u>23.5</u>	<u>27.1</u>	<u>31.3</u>	<u>34.3</u>	<u>34.7</u>
Gauge Height Mile 5 (feet)	3.40 ¹											
Discharge Mile 5 (c.f.s.)	77											
Air Temperature (°C.)	High - 17.1 Low - 14.3											
Water Temperature (°C.)	21.0	19.0	18.5	19.0	18.5	19.5	20.0	16.5	16.0	16.5	16.5	16.5
Conductivity (x 10 ⁻⁶ ohms ⁻¹ cm ⁻¹)												
Alkalinity (mg l ⁻¹)	266	266	266	268	266	270	270	268	266	260	256	252
pH	8.05	8.10	8.05	7.90	7.90	7.90	7.90	8.05	7.95	8.00	8.05	8.05
Turbidity (units)	6	6	4	3	4	6	6	4	3	4	4	5
Dissolved Oxygen (mg l ⁻¹)	6.6	7.5	8.0	6.5	6.0	6.2	6.2	7.9	7.1	7.1	8.6	8.7
Total Phosphorus (µg l ⁻¹)	96	62	53	61	66	137	110	66	68	85	18	15
Nitrogen•NH ₄ (mg l ⁻¹)												
Nitrogen•NO ₂ + NO ₃ (mg l ⁻¹)												

¹Data from U. S. Geological Survey, Lansing, Michigan Office

²Data from U. S. Weather Bureau, East Lansing, Michigan Office

Physical and chemical characteristics of the Red Cedar River - July 10, 1956

Miles from Mouth 4.5 7.8 10.3 13.6 16.6 18.3 18.8 23.5 27.1 31.3 34.3 34.7

Gauge Height Mile 5 (feet) 3.38¹

Discharge Mile 5 (c.f.s.) 72

Air Temperature (°C.) High - 27.3 Low - 12.6²

Water Temperature (°C.) 20.5 20.5 19.5 21.0 20.5 19.5

Conductivity
(x 10⁻⁶ ohms⁻¹ cm⁻¹)

Alkalinity (mg l⁻¹) 266 266 266 266 268 270 268 266 266 266 260

pH 8.00 8.00 8.05 8.05 8.00 7.95 7.95 8.15 8.05 8.25 8.20

Turbidity (units) 9 6 2 4 4 5 9 5 4 4 4

Dissolved Oxygen (mg l⁻¹)

Total Phosphorus (µg l⁻¹) 68 38 49 44 37 158 167 30 47 57 13

Nitrogen·NH₄ (mg l⁻¹)

Nitrogen·NO₂ + NO₃ (mg l⁻¹)

¹Data from U. S. Geological Survey, Lansing, Michigan Office

²Data from U. S. Weather Bureau, East Lansing, Michigan Office

Physical and chemical characteristics of the Red Cedar River - August 10, 1956

Miles from Mouth	4.5	7.8	10.3	13.6	16.6	18.3	18.8	23.5	27.1	31.3	34.3	34.7
Gauge Height Mile 5 (feet)	4.29 ¹											
Discharge Mile 5 (c.f.s.)	363											
Air Temperature (°C.)		High - 28.8	Low - 16.1									
Water Temperature (°C.)	23.5	24.0	24.0	22.0	21.5	22.0	22.5	20.5	22.0	20.0	20.0	19.0
Conductivity (x 10 ⁻⁶ ohms ⁻¹ cm ⁻¹)	372	415	425	468	430	425	408	405	451	469	469	440
Alkalinity (mg l ⁻¹)	170	182	190	210	194	190	190	208	208	214	218	202
pH	7.85	7.75	7.70	7.65	7.65	7.60	7.60	7.50	7.40	7.30	7.30	7.25
Turbidity (units)	33	35	39	28	33	50	43	29	21	16	17	18
Dissolved Oxygen (mg l ⁻¹)	5.9	5.8	5.7	5.9	6.0	5.9	5.3	5.1	4.2	3.4	3.7	5.1
Total Phosphorus (µg l ⁻¹)	204	137	196	84	110	133	115	122	91	97	66	54
Nitrogen•NH ₄ (mg l ⁻¹)												
Nitrogen•NO ₂ + NO ₃ (mg l ⁻¹)												

¹Data from U. S. Geological Survey, Lansing, Michigan Office

²Data from U. S. Weather Bureau, East Lansing, Michigan Office

Physical and chemical characteristics of the Red Cedar River - August 17, 1956

Miles from Mouth 4.5 7.8 10.3 13.6 16.6 18.3 18.8 23.5 27.1 31.3 34.3 34.7

Gauge Height Mile 5 (feet) 3.45¹

Discharge Mile 5 (c.f.s.) 90

Air Temperature (°C.) High - 30.4 Low - 18.2²

Water Temperature (°C.) 24.0 24.5 25.0 25.0 24.0 24.5 22.5 21.5 23.0 21.0 19.5

Conductivity
(x 10⁻⁶ ohms⁻¹ cm⁻¹) 673 540 540 508 585 575 568 597 527 546 513

Alkalinity (mg l⁻¹) 274 278 279 279 283 280 290 288 294 300 262

pH 8.05 8.05 8.30 8.45 8.10 8.05 8.20 7.95 7.50 7.50 7.50

Turbidity (units) 9 7 6 5 6 6 6 4 4 5 4

Dissolved Oxygen (mg l⁻¹) 5.7 6.5 6.5 6.1 5.8 5.0 6.4 5.6 4.7 5.3 6.7

Total Phosphorus (µg l⁻¹) 110 86 91 64 183 160 96 68 93 25 20

Nitrogen-NH₄ (mg l⁻¹) 0.65 0.03 0.03 0.03 0.22 0.24 0.04 0.04 0.02 0.04 0.04

Nitrogen-NO₂ + NO₃ (mg l⁻¹) 0.17 0.25 0.16 0.13 0.20 0.23 0.14 0.16 0.14 0.08 0.08

¹Data from U. S. Geological Survey, Lansing, Michigan Office

²Data from U. S. Weather Bureau, East Lansing, Michigan Office

Physical and chemical characteristics of the Red Cedar River - August 27, 1956

Miles from Mouth 4.5 7.8 10.3 13.6 16.6 18.3 18.8 23.5 27.1 31.3 34.3 34.7

Gauge Height Mile 5 (feet) 3.35¹

Discharge Mile 5 (c.f.s.) 65

Air Temperature (°C.) High - 31.5 Low - 16.6²

Water Temperature (°C.) 21.5 22.0 22.5 23.5 21.0 21.0 20.0 20.0 20.0 20.0 17.5 17.0

Conductivity
(x 10⁻⁴ ohms⁻¹ cm⁻¹) 545 545 527 533 571 563 549 549 549 549 503 497

Alkalinity (mg l⁻¹) 278 278 280 280 284 278 290 280 280 280 280 264

pH 8.60 8.10 8.45 8.25 8.10 7.90 7.70 7.65 7.60 7.60 7.60 7.60

Turbidity (units) 8 3 4 4 6 4 4 4 4 4 4 9

Dissolved Oxygen (mg l⁻¹) 7.1 8.0 8.1 7.5 6.5 6.6 7.6 7.0 6.3 7.0 8.1

Total Phosphorus (µg l⁻¹) 86 61 64 62 267 237 54 66 99 19 8

Nitrogen·NH₄ (mg l⁻¹) 0.04 0.0 0.0 0.04 0.51 0.25 0.0 0.0 0.0 0.0 0.0

Nitrogen·NO₂ + NO₃ (mg l⁻¹) 0.26 0.25 0.27 0.21 0.35 0.04 0.36 0.34 0.42 0.27 0.31

¹Data from U. S. Geological Survey, Lansing, Michigan Office

²Data from U. S. Weather Bureau, East Lansing, Michigan Office

Physical and chemical characteristics of the Red Cedar River - September 20, 1956

Miles from Mouth	4.5	7.8	10.3	13.6	16.6	18.3	18.8	23.5	27.1	31.3	34.3	34.7
Gauge Height Mile 5 (feet)	3.24 ¹											
Discharge Mile 5 (c.f.s.)	40											
Air Temperature (°C.)												
Water Temperature (°C.)	13.5	12.0	12.5	12.0	13.0	13.5	13.5	10.0	10.0	10.5	7.5	7.5
Conductivity (x 10 ⁻⁶ ohms ⁻¹ cm ⁻¹)	587	529	522	529	523	472	530	517	466	421	485	485
Alkalinity (mg l ⁻¹)	266	272	268	270	274	274	280	272	274	280	274	235
pH	7.75	7.80	7.75	7.65	7.60	7.55	7.50	7.45	7.40	7.15	7.15	7.15
Turbidity (units)	6	2	2	2	4	6	6	6	4	4	5	6
Dissolved Oxygen (mg l ⁻¹)	9.7	11.6	12.1	10.2	9.6	7.5	7.8	10.0	8.9	6.3	9.0	9.1
Total Phosphorus (µg l ⁻¹)	110	42	61	61	47	442	248	50	56	140	13	14
Nitrogen-NH ₄ (mg l ⁻¹)	0.13	0.0	0.0	0.0	0.04	0.70	0.32	0.0	0.0	0.0	0.0	0.0
Nitrogen-NO ₂ + NO ₃ (mg l ⁻¹)	0.26	0.25	0.25	0.29	0.29	0.24	0.22	0.19	0.18	0.25	0.15	0.11

¹Data from U. S. Geological Survey, Lansing, Michigan Office

²Data from U. S. Weather Bureau, East Lansing, Michigan Office

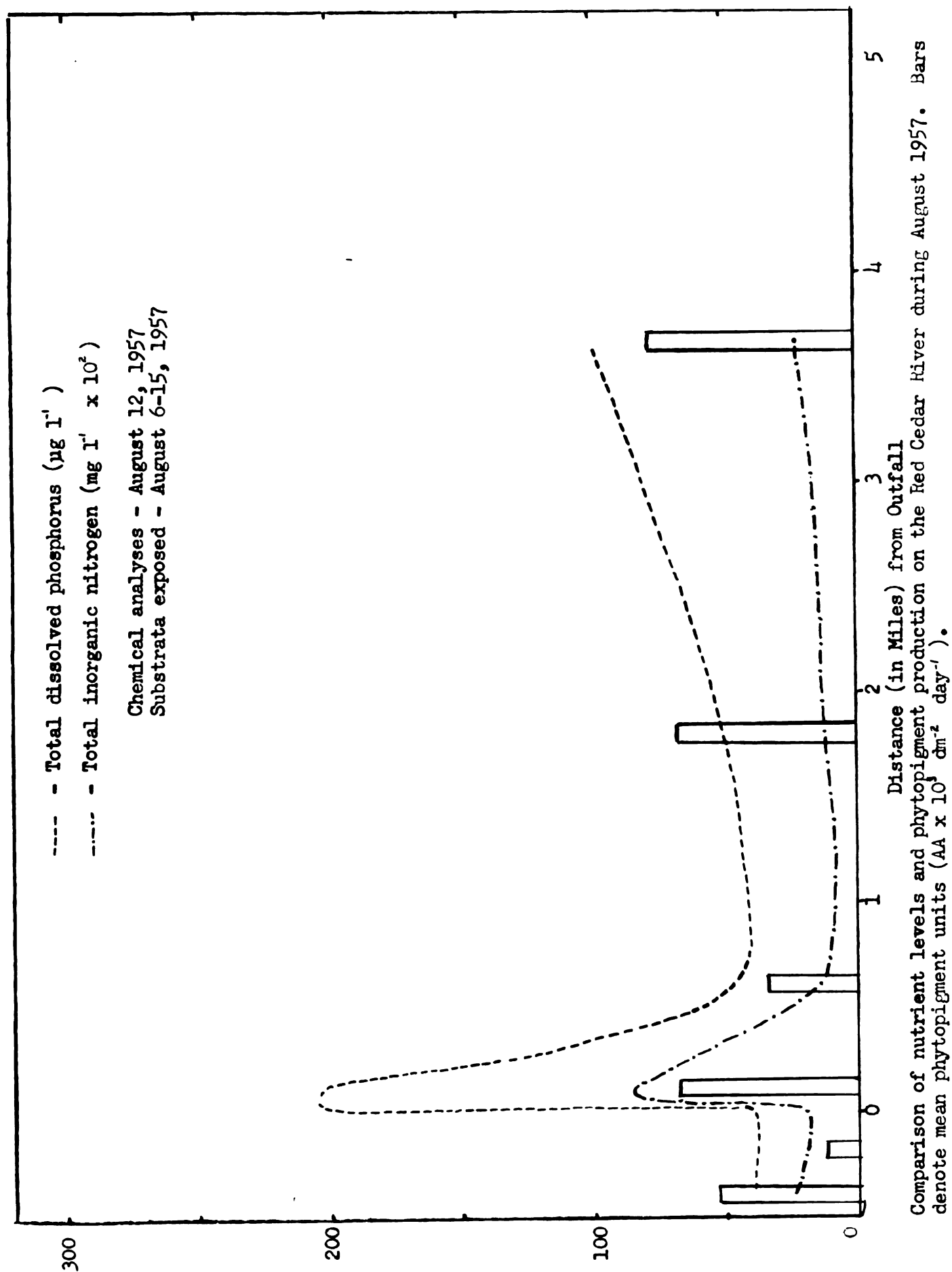
Physical and chemical characteristics of the Red Cedar River - September 30, 1956

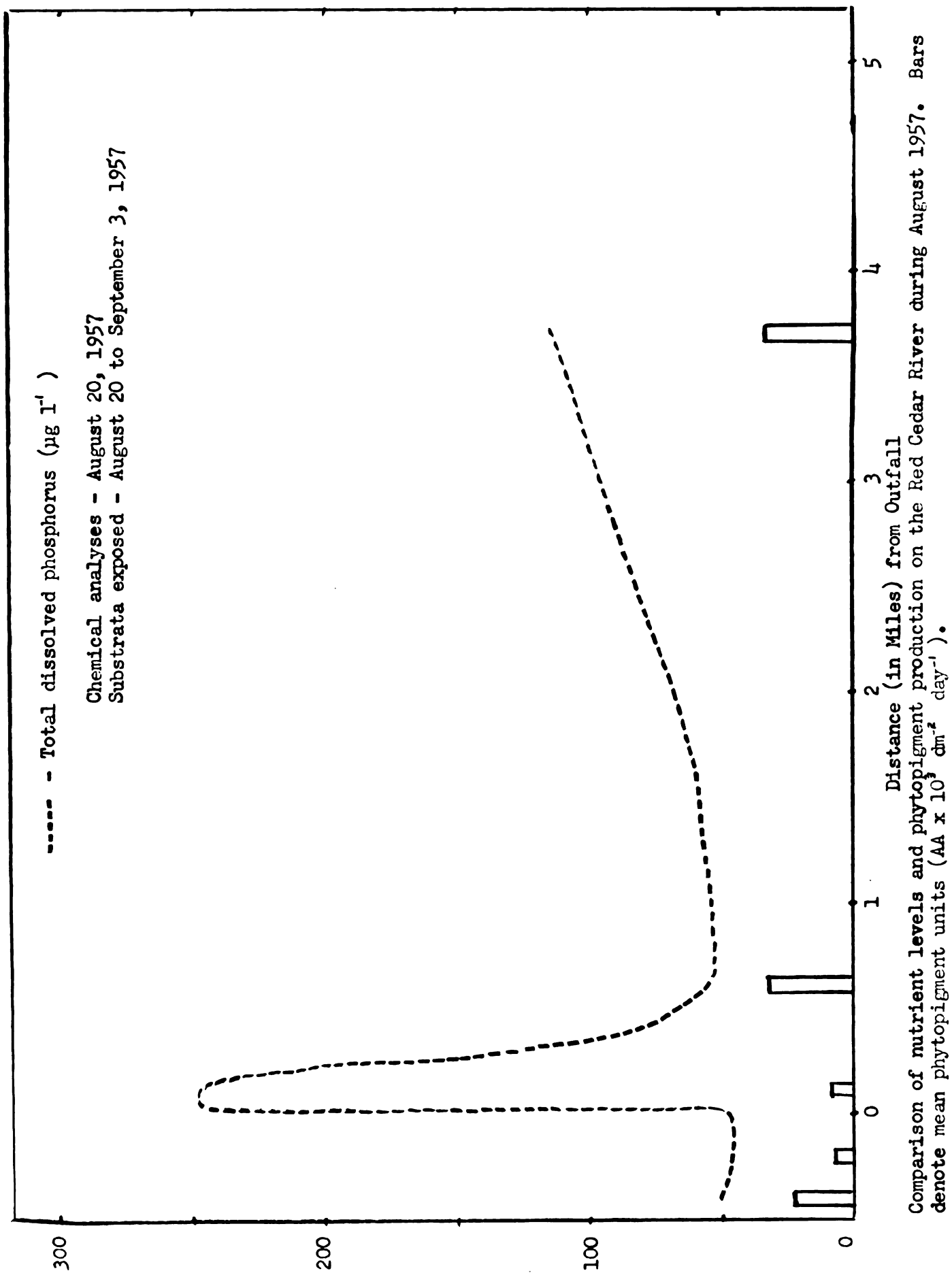
<u>Miles from Mouth</u>	<u>4.5</u>	<u>7.8</u>	<u>10.3</u>	<u>13.6</u>	<u>16.6</u>	<u>18.3</u>	<u>18.8</u>	<u>23.5</u>	<u>27.1</u>	<u>31.3</u>	<u>34.3</u>	<u>34.7</u>
Gauge Height Mile 5 (feet)	3.22 ¹											
Discharge Mile 5 (c.f.s.)	36											
Air Temperature (°C.)	High - 16.6 Low - 6.0 ²											
Water Temperature (°C.)	17.0	15.0	15.5	16.0	15.0	17.0	16.0	14.5	14.5	14.0	13.0	12.5
Conductivity (x 10 ⁻⁶ ohms ⁻¹ cm ⁻¹)	519	512	514	520	514	506	520	514	538	557	488	475
Alkalinity (mg l ⁻¹)	258	256	258	260	258	272	268	268	270	274	272	276
pH	7.85	8.25	8.20	8.05	8.00	7.80	7.50	7.65	7.65	7.65	7.65	7.80
Turbidity (units)	-											
Dissolved Oxygen (mg l ⁻¹)	9.2	10.5	10.1	8.8	9.2	8.3	8.8	8.8	8.1	4.5	7.0	7.5
Total Phosphorus (ug l ⁻¹)	115	39	44	70	71	125	125	61	63	177	19	13
Nitrogen•NH ₄ (mg l ⁻¹)	0.13	0.0	0.0	0.0	0.03	0.35	0.30	0.0	0.0	0.10	0.03	0.05
Nitrogen•NO ₂ + NO ₃ (mg l ⁻¹)	0.13	0.13	0.13	0.15	0.14	0.16	0.13	0.18	0.18	0.22	0.04	0.04

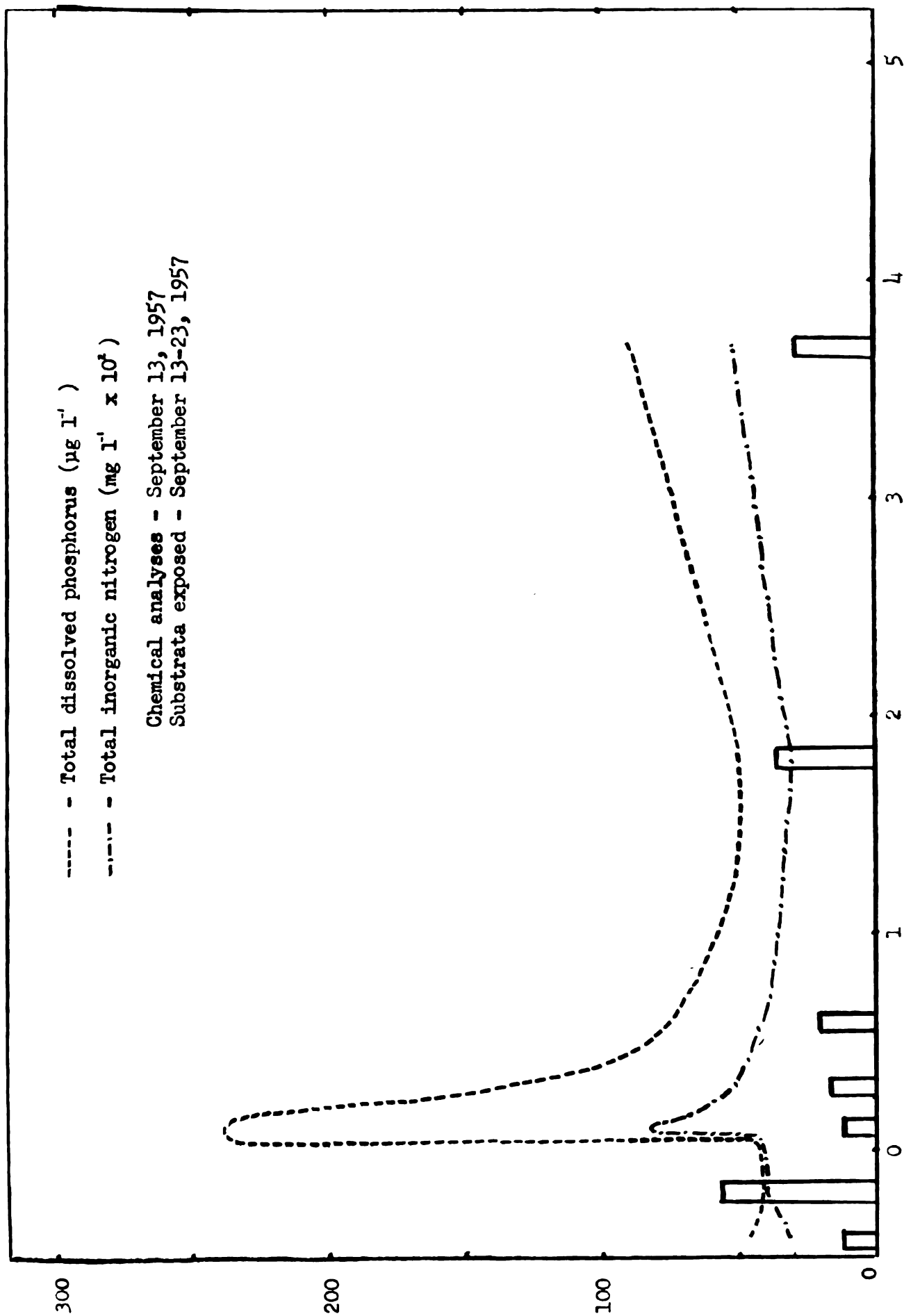
¹Data from U. S. Geological Survey, Lansing, Michigan Office

²Data from U. S. Weather Bureau, East Lansing, Michigan Office

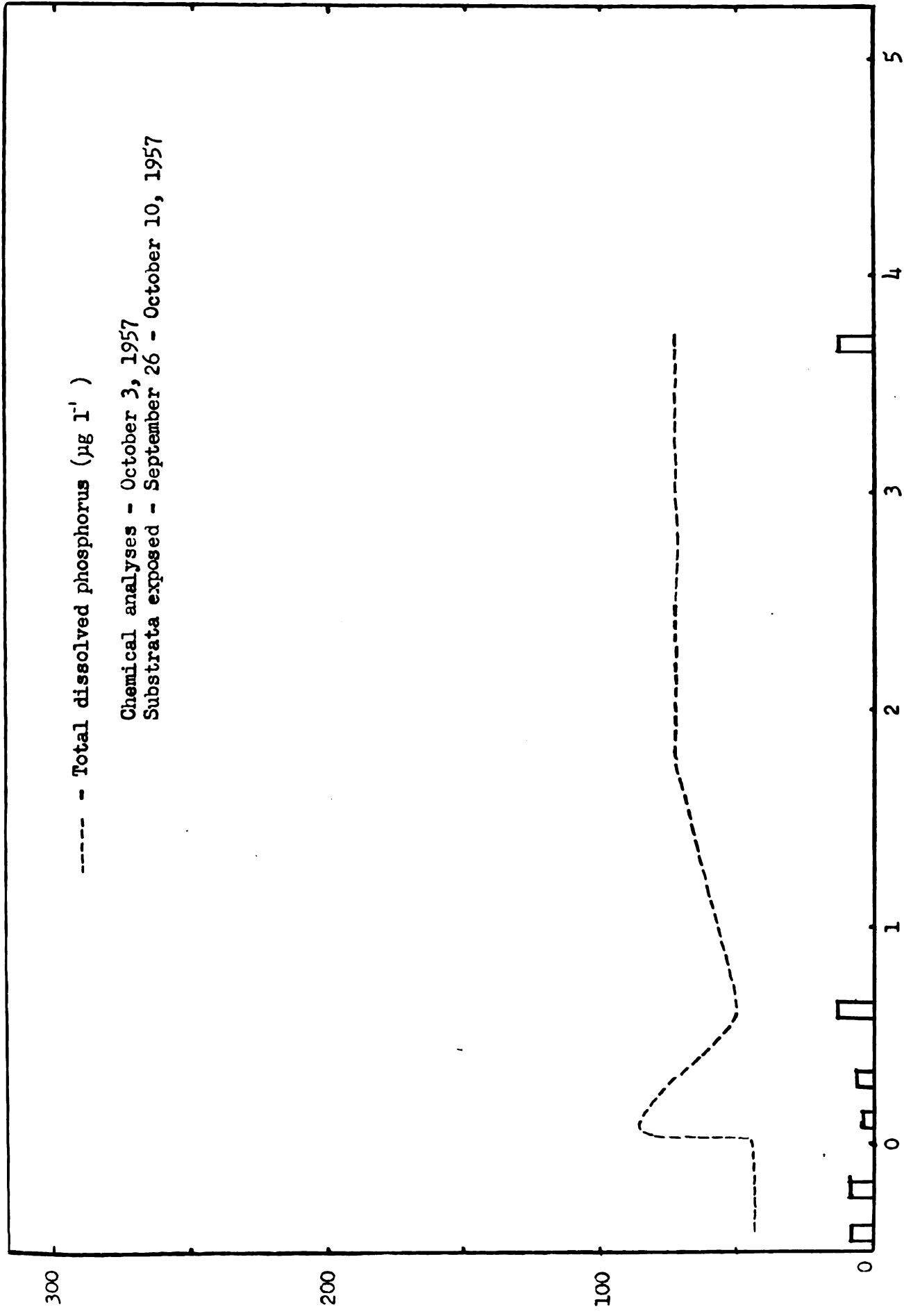
APPENDIX C







Comparison of nutrient levels and phytoplankton production on the Red Cedar River during September 1957. Bars denote mean phytoplankton production ($\text{AA} \times 10^3 \text{ cm}^{-2} \text{ day}^{-1}$).



Comparison of nutrient levels and phytoplankton production on the Red Cedar River during October 1957. Bars denote mean phytoplankton units ($\text{AA} \times 10^3 \text{ cm}^{-2} \text{ day}^{-1}$).

----- Total dissolved phosphorus ($\mu\text{g l}^{-1}$)

- - - - - Total inorganic nitrogen ($\text{mg l}^{-1} \times 10^4$)

Chemical analyses - October 31, 1957
Substrata exposed - October 21 to November 4, 1957

300

200

100

0

1

2

3

4

5

Distance (in Miles) from Outfall

Comparison of nutrient levels and phytoplankton production on the Red Cedar River during October 1957. Bars denote mean phytoplankton units ($\text{AA} \times 10^3 \text{ dm}^{-2} \text{ day}^{-1}$).

300

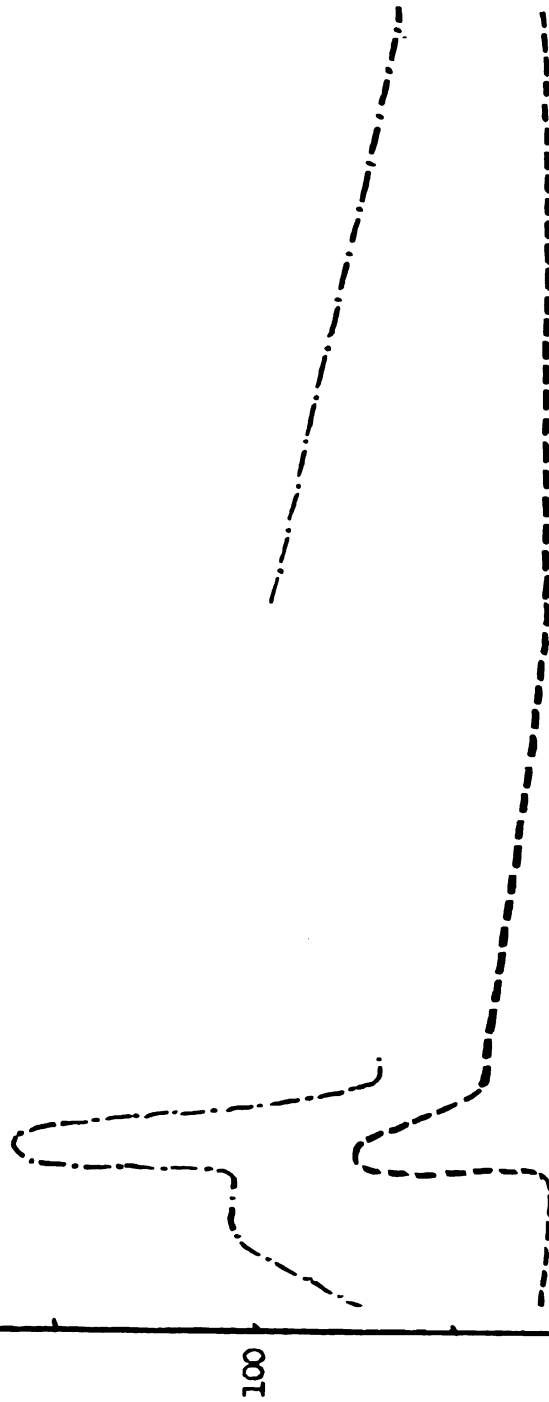
200

100

0

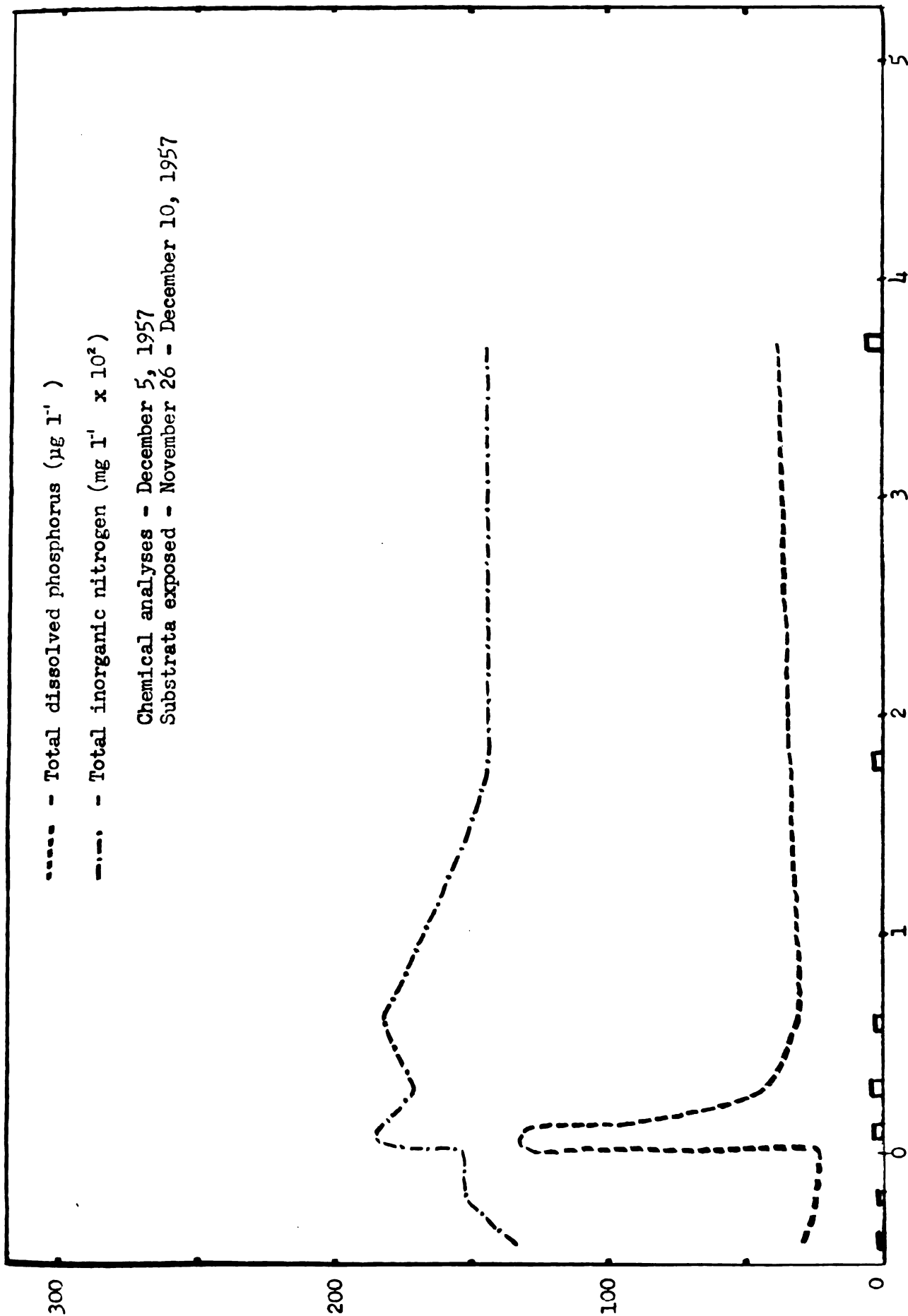
----- Total dissolved phosphorus ($\mu\text{g l}^{-1}$)- - - - - Total inorganic nitrogen ($\text{mg l}^{-1} \times 10^2$)

Chemical analyses - November 19, 1957
 Substrata exposed - November 7-21, 1957



Distance (in Miles) from Outfall

Comparison of nutrient levels and phytopigment production on the Red Cedar River during November 1957. Bars denote mean phytopigment units ($\text{AA} \times 10^3 \text{ dm}^{-2} \text{ day}^{-1}$).



Comparison of nutrient levels and phytoplankton production on the Red Cedar River during December 1957. Bars denote mean phytoplankton units ($\text{AA} \times 10^3 \text{ dm}^{-2} \text{ day}^{-1}$).

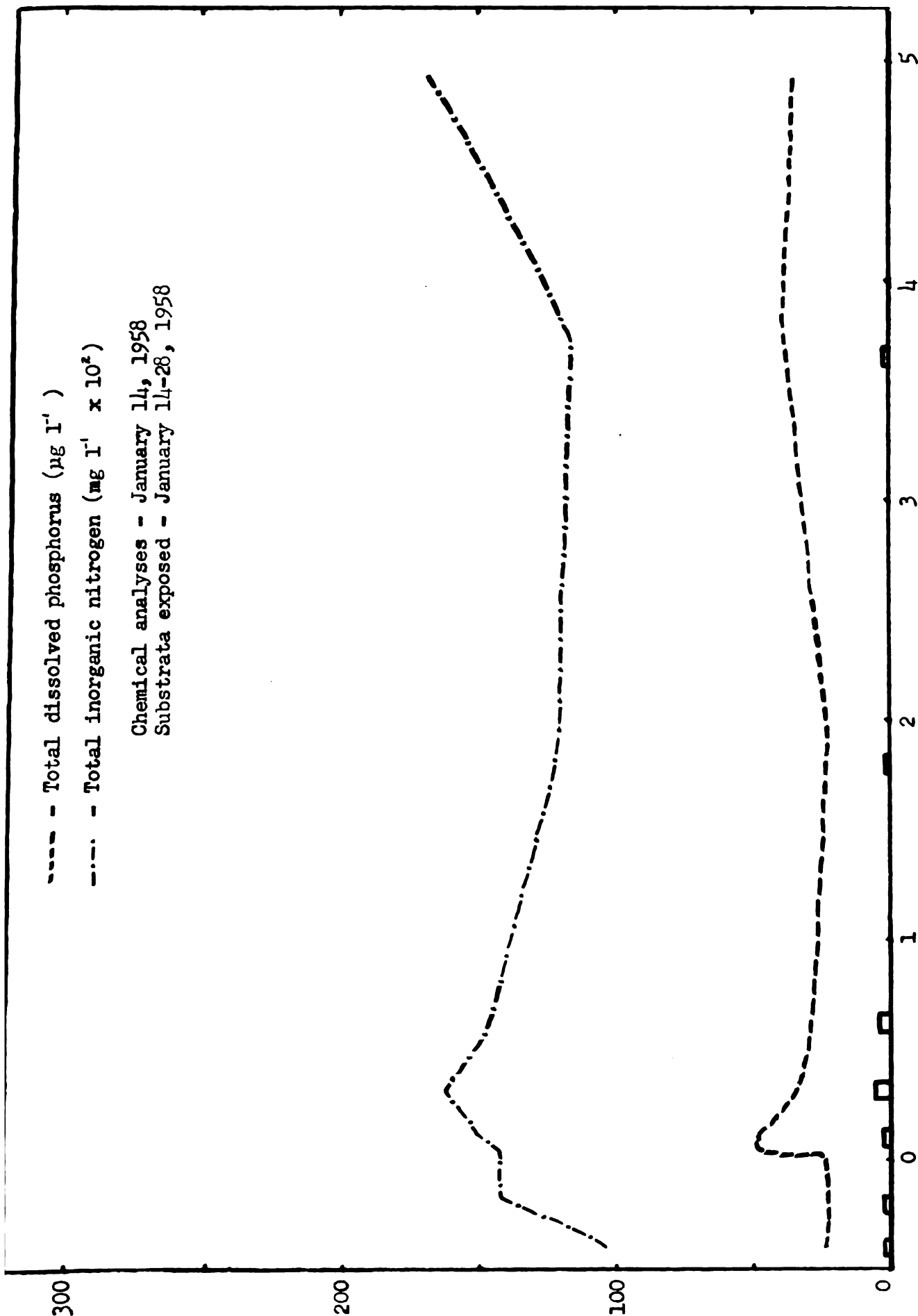
Substrata exposed - December 17, 1957 to January 7, 1958

Q 8

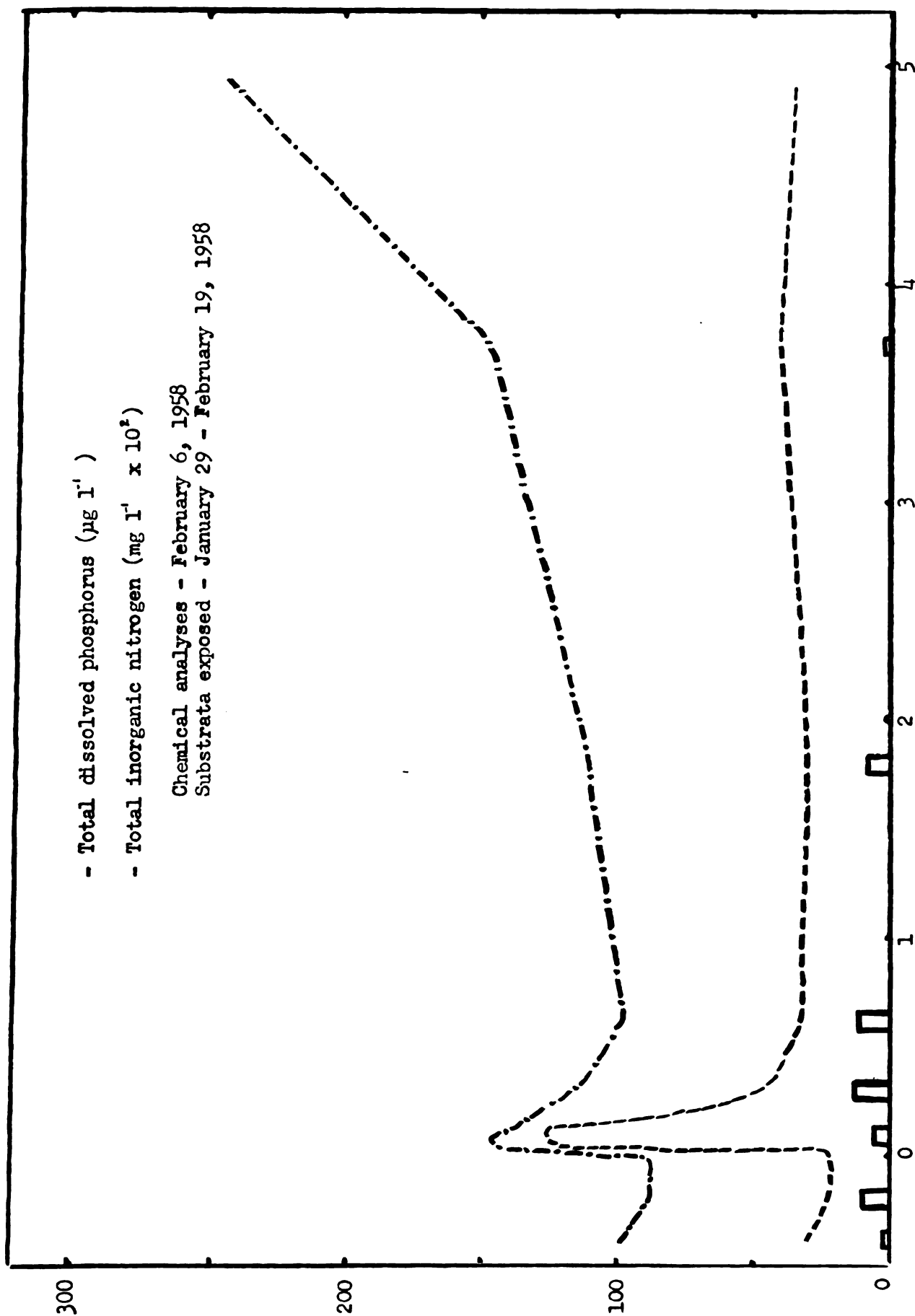


Phytoplankton production on the Red Cedar River during December 1957. Bars denote mean phytoplankton units (AA x 10³ dm⁻² day⁻¹).

- - - - - Total dissolved phosphorus ($\mu\text{g l}^{-1}$)
 - - Total inorganic nitrogen ($\text{mg l}^{-1} \times 10^2$)
 Chemical analyses - January 14, 1958
 Substrata exposed - January 14-28, 1958

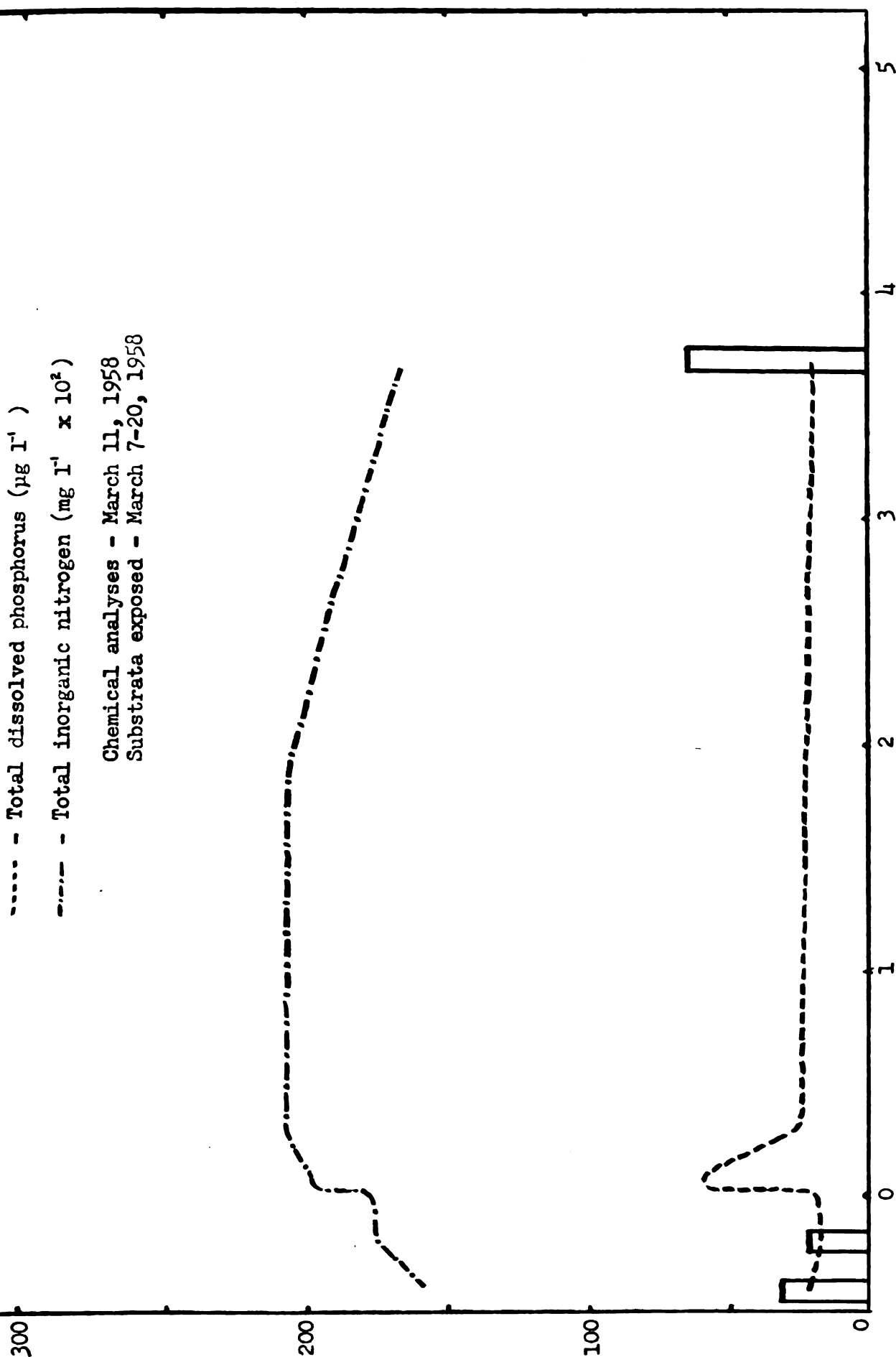


Comparison of nutrient levels and phytoplankton production on the Red Cedar River during January 1958. Bars denote mean phytoplankton units ($\text{AA} \times 10^3 \text{ dm}^{-2} \text{ day}^{-1}$).



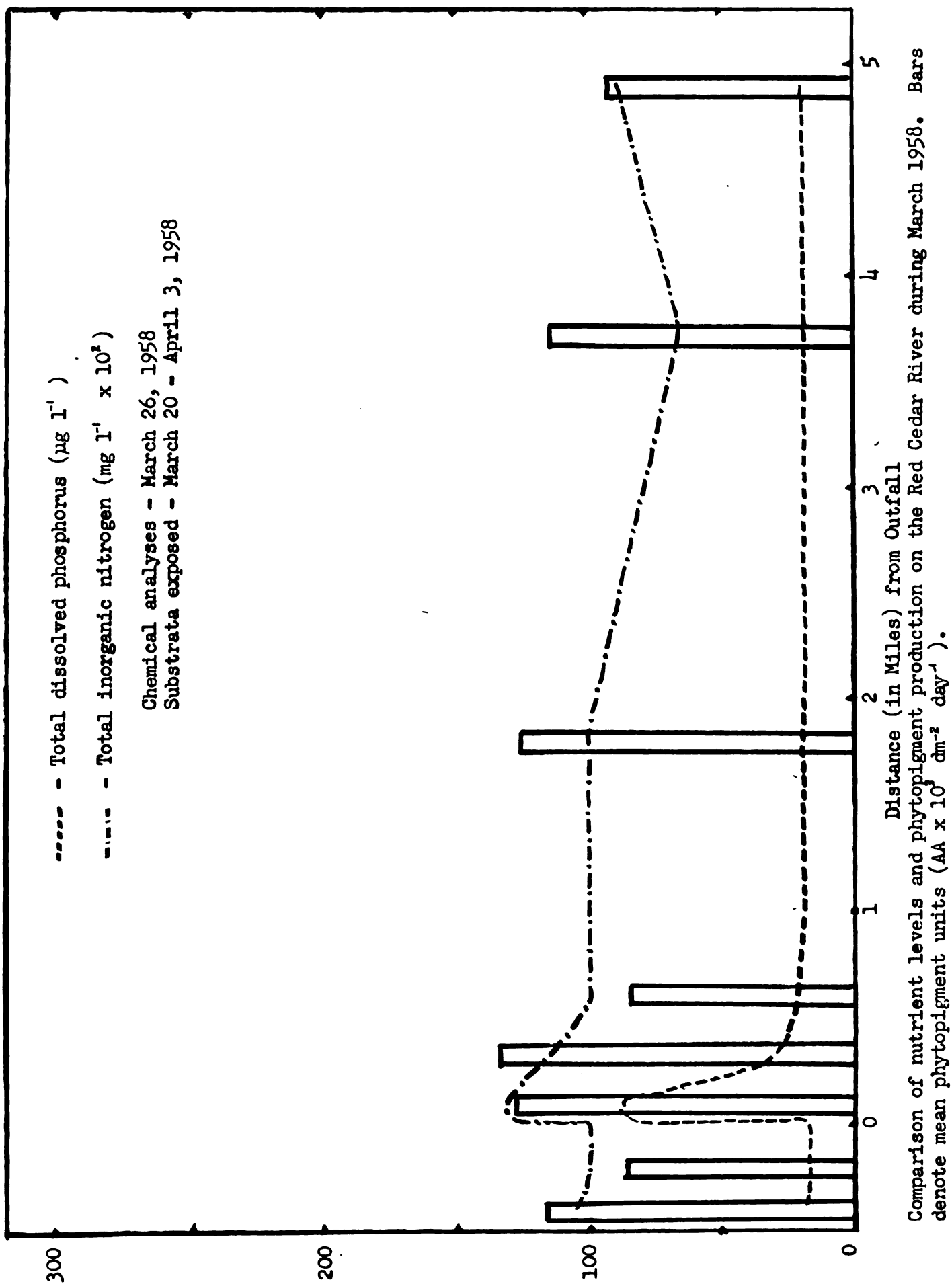
..... - Total dissolved phosphorus ($\mu\text{g l}^{-1}$)
 - - - - - Total inorganic nitrogen ($\text{mg l}^{-1} \times 10^2$)

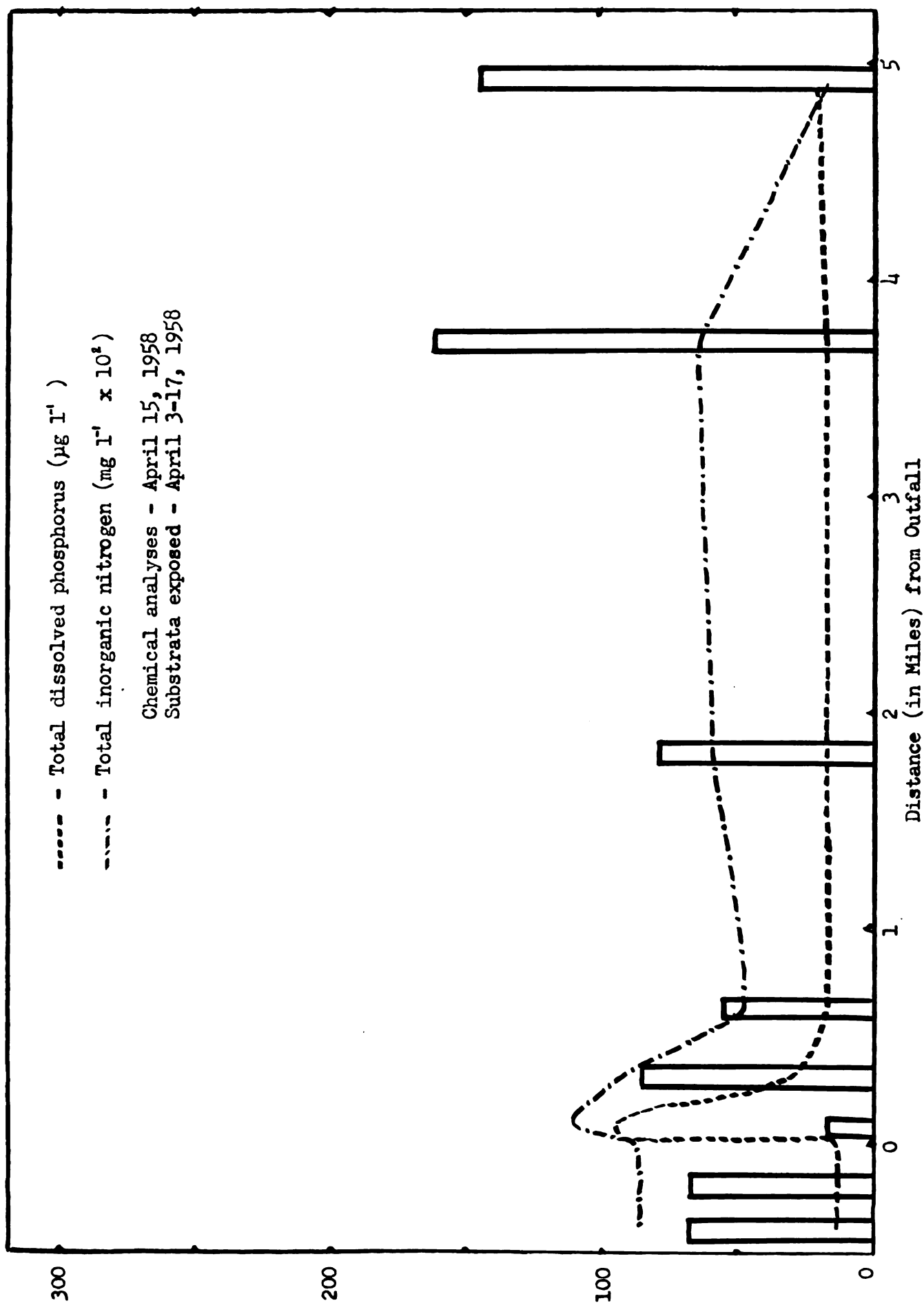
Chemical analyses - March 11, 1958
 Substrata exposed - March 7-20, 1958



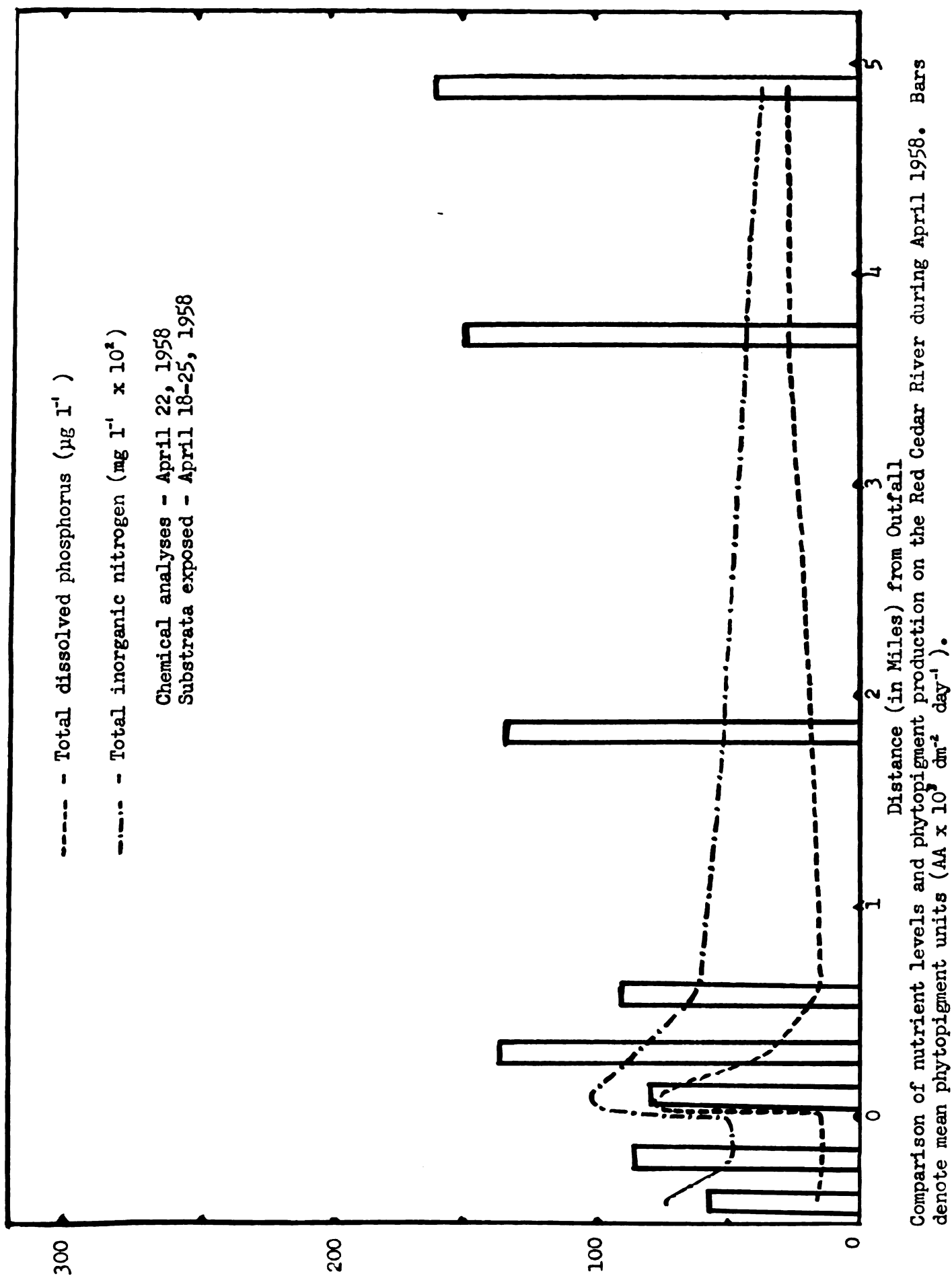
Distance (in Miles) from Outfall

Comparison of nutrient levels and phytoplankton production on the Red Cedar River during March 1958. Bars denote mean phytoplankton units ($\text{AA} \times 10^3 \text{ cm}^{-2} \text{ day}^{-1}$).



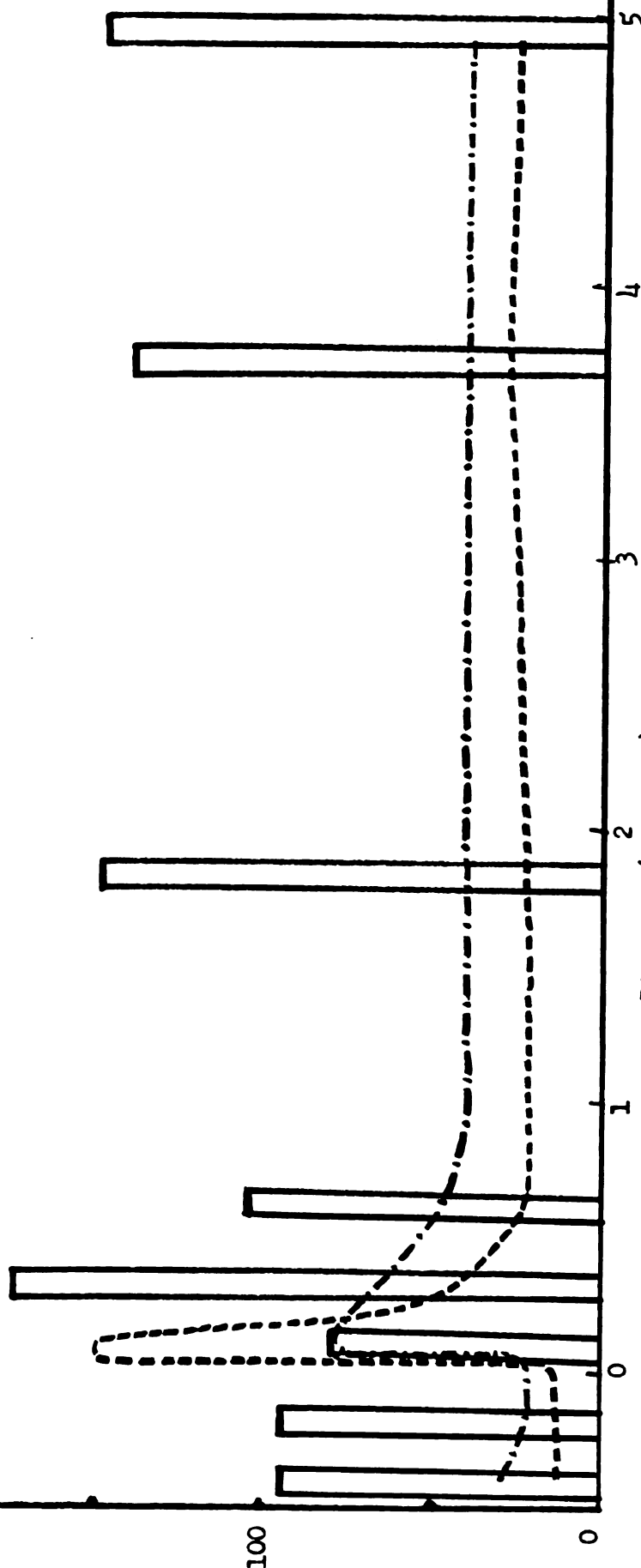


Comparison of nutrient levels and phytoplankton production on the Red Cedar River during April 1958. Bars denote mean phytoplankton units ($\text{AA} \times 10^3 \text{ dm}^{-2} \text{ day}^{-1}$).

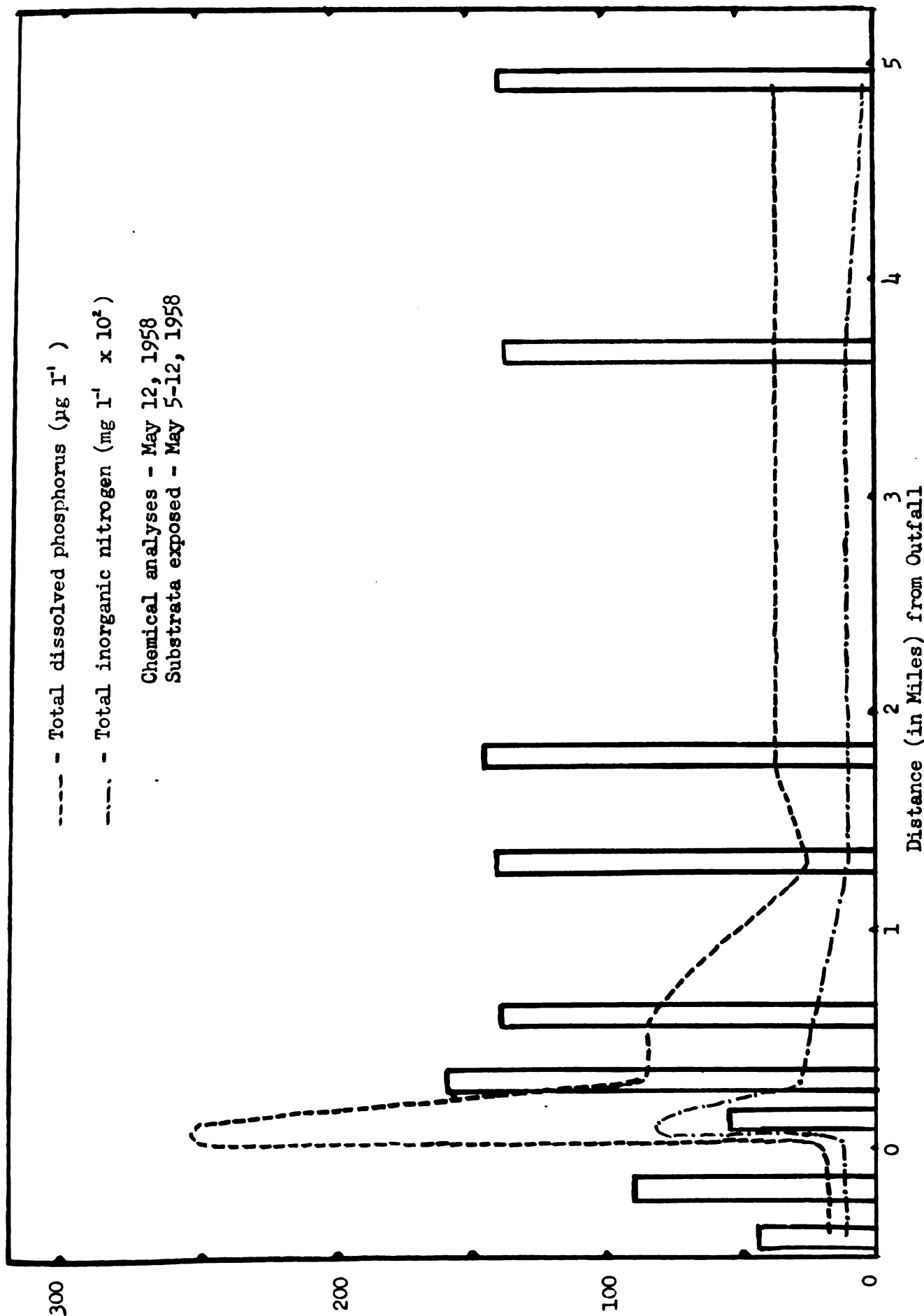


----- Total dissolved phosphorus ($\mu\text{g l}^{-1}$)
 - - - - - Total inorganic nitrogen ($\text{mg l}^{-1} \times 10^2$)

Chemical analyses - April 30, 1958
 Substrata exposed - April 25 - May 5, 1958

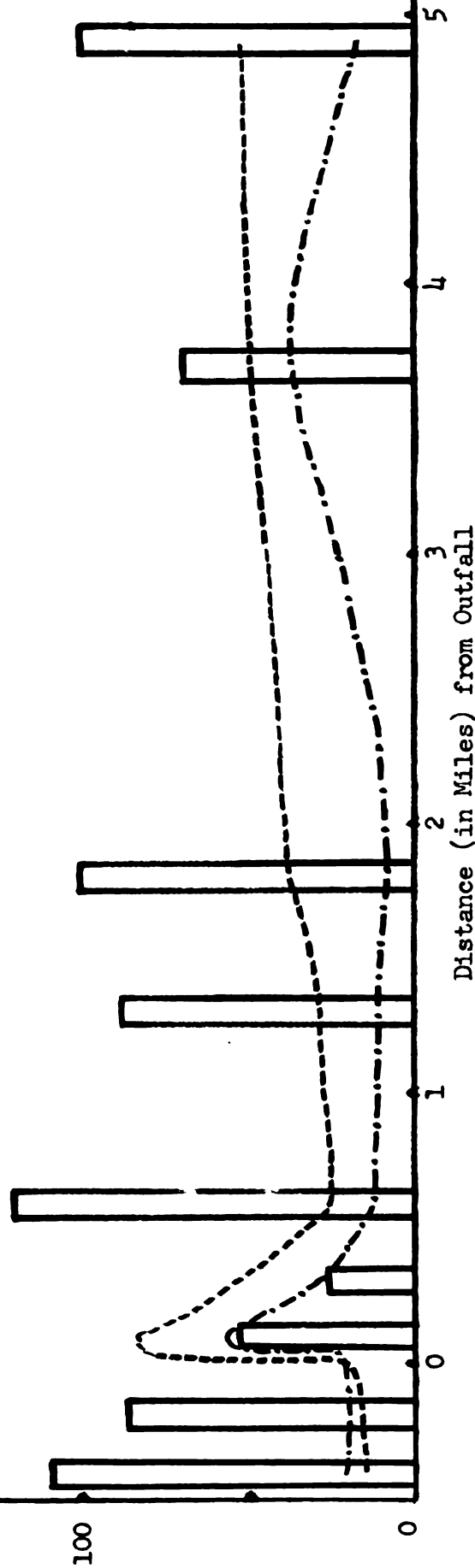


Comparison of nutrient levels and phytoplankton production on the Red Cedar River during April 1958. Bars denote mean phytoplankton units ($\text{AA} \times 10^3 \text{ dm}^{-2} \text{ day}^{-1}$).

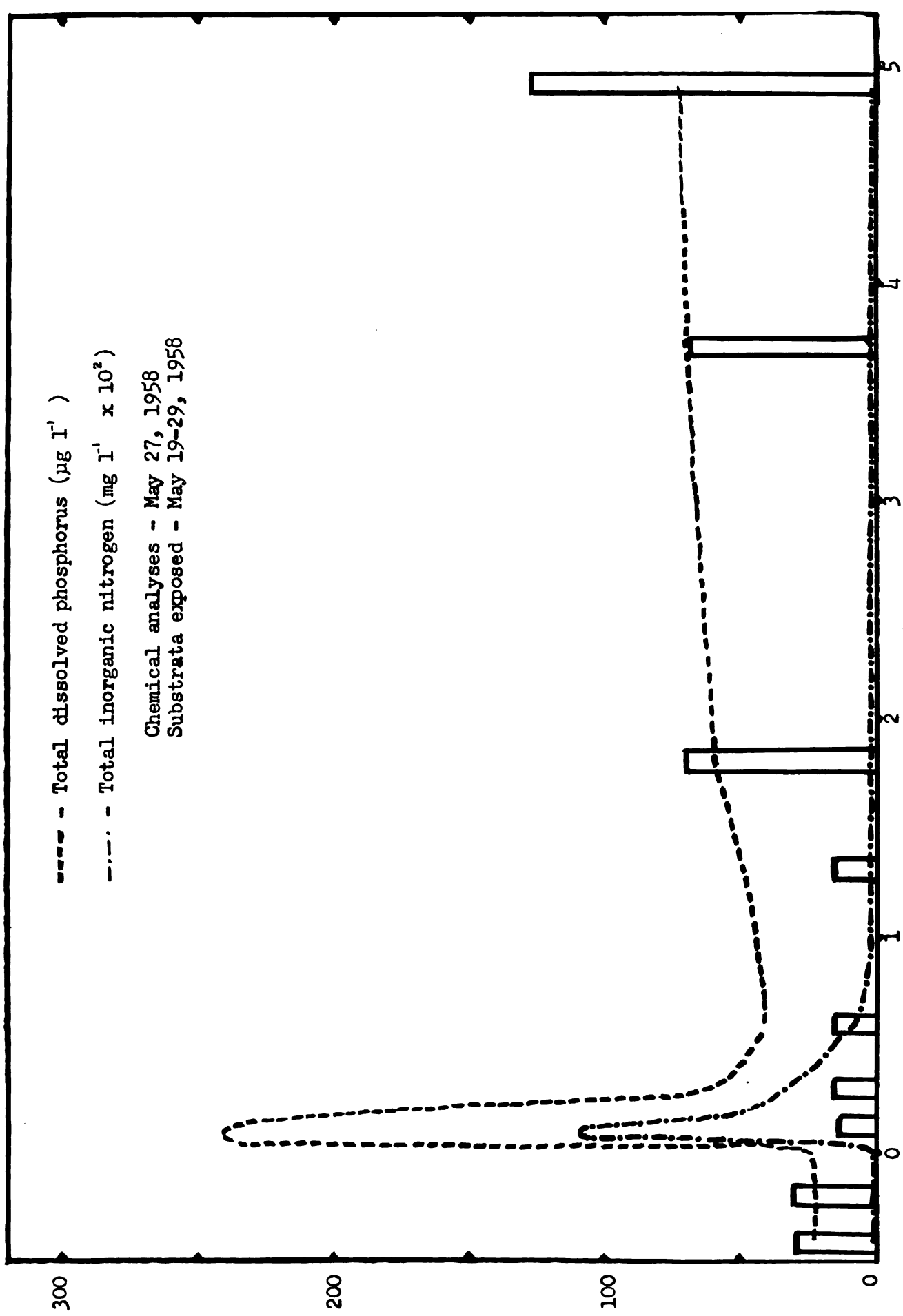


Comparison of nutrient levels and phytoplankton production on the Red Cedar River during May 1958. Bars denote mean phytoplankton units ($\text{AA} \times 10^2 \text{ dm}^{-2} \text{ day}^{-1}$).

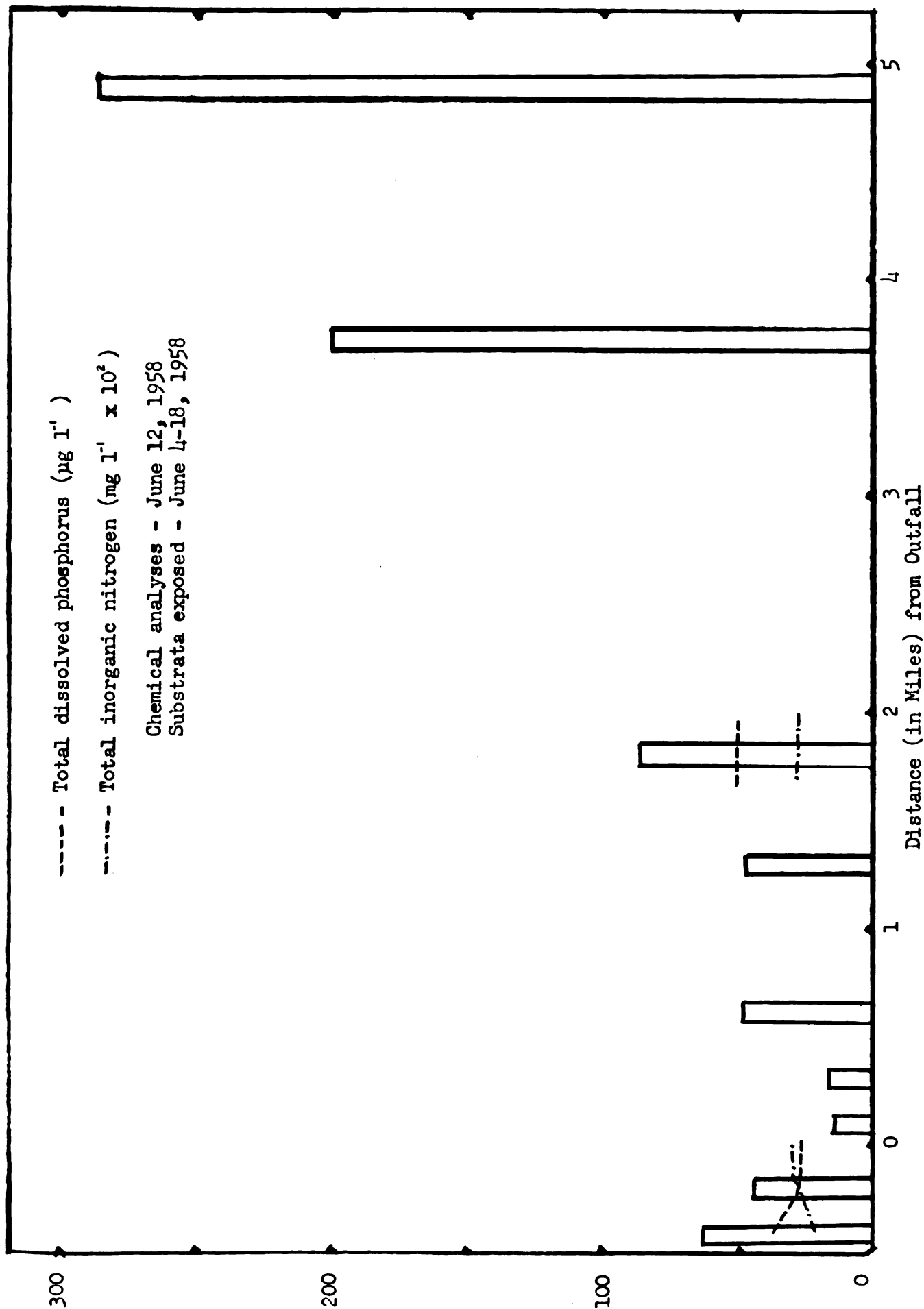
- - - - - Total dissolved phosphorus ($\mu\text{g l}^{-1}$)
 - - - - - Total inorganic nitrogen ($\text{mg l}^{-1} \times 10^2$)
 Chemical analyses - May 15, 1958
 Substrata exposed - May 12-19, 1958



Comparison of nutrient levels and phytopigment production on the Red Cedar River during May 1958. Bars denote mean phytopigment units ($\text{AA} \times 10^3 \text{ dm}^{-2} \text{ day}^{-1}$).

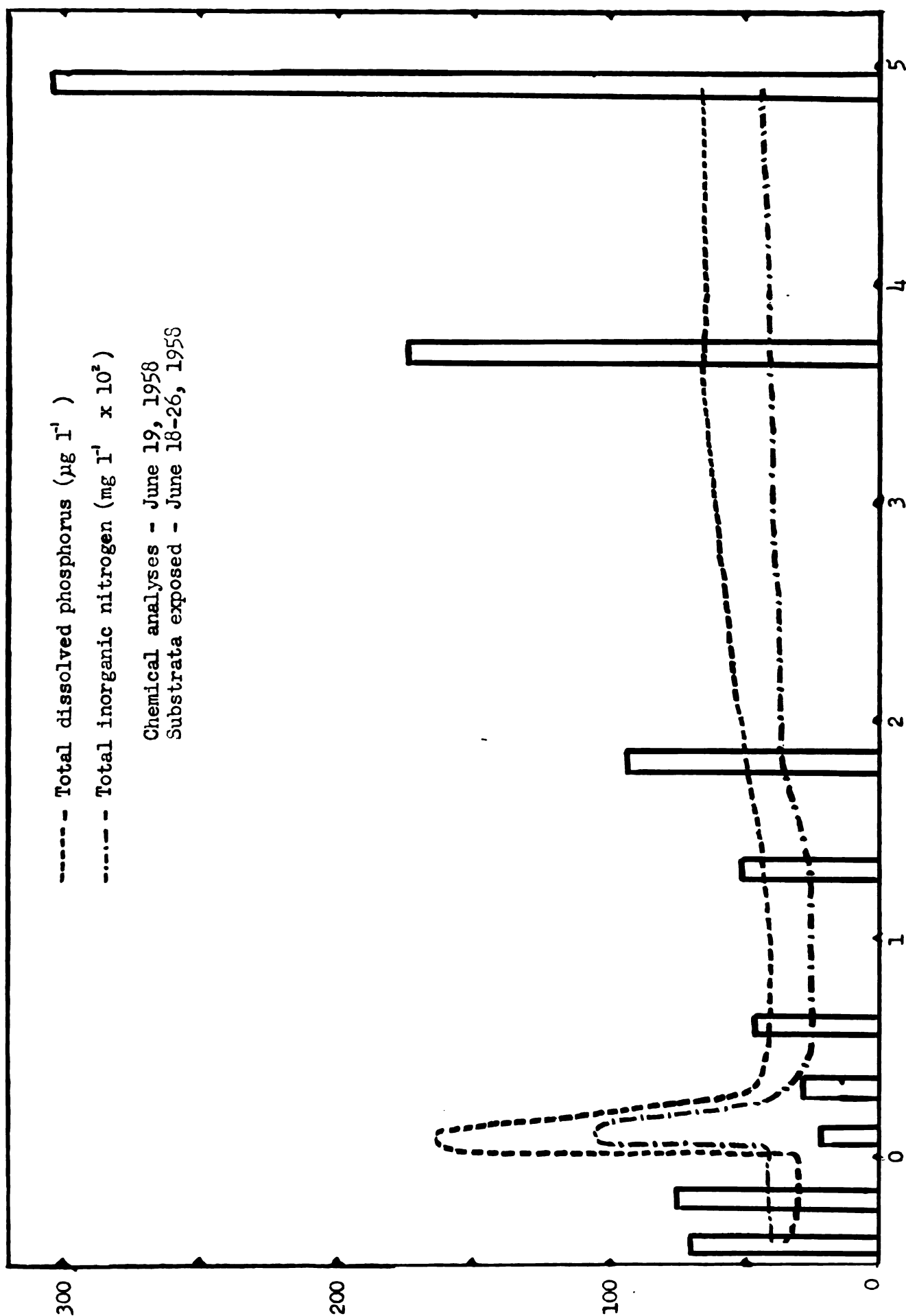


Comparison of nutrient levels and phytoplankton production on the Red Cedar River during May 1958. Bars denote mean phytoplankton units ($\text{AA} \times 10^3 \text{ cm}^{-2} \text{ day}^{-1}$).



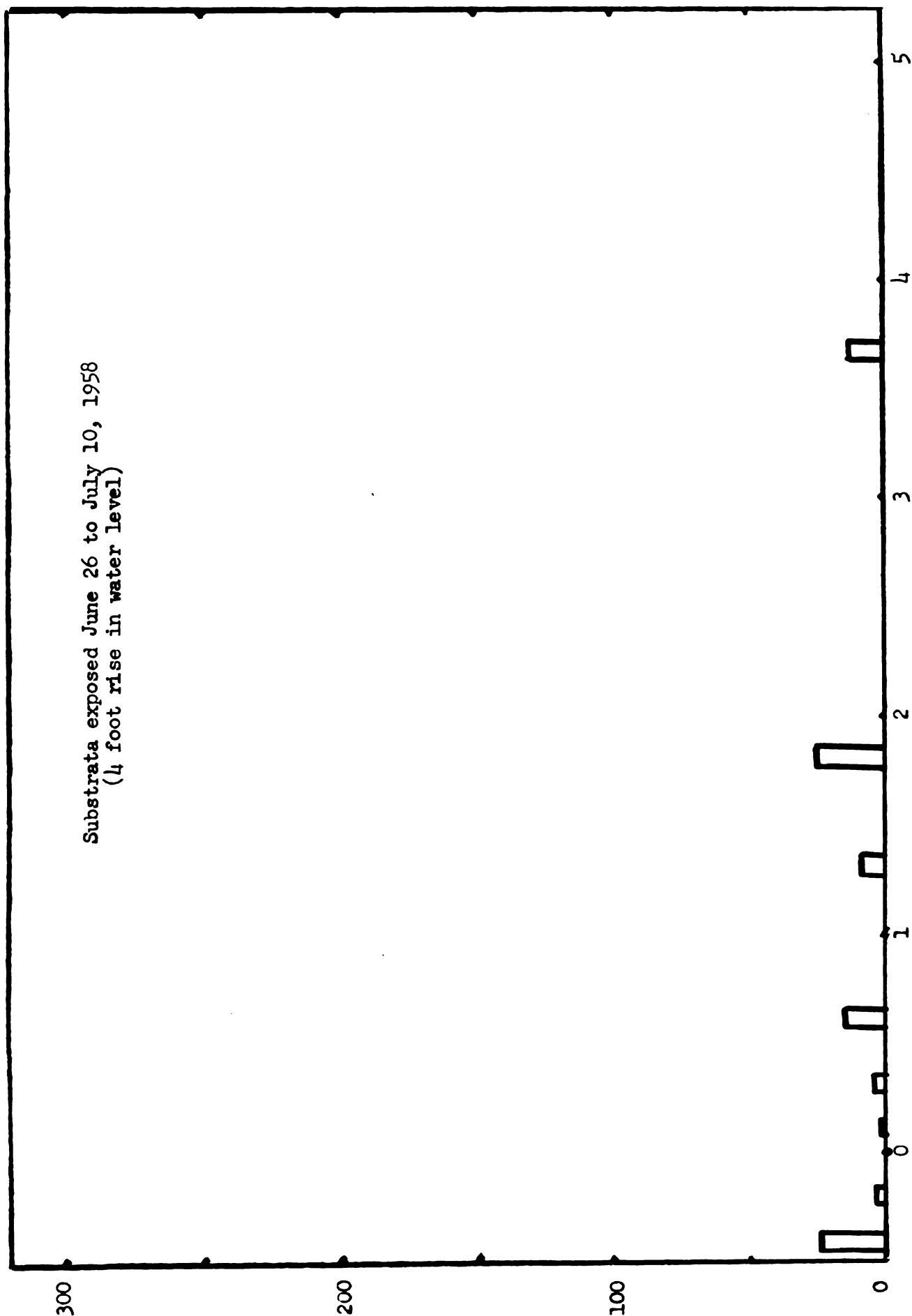
Distance (in Miles) from Outfall

Comparison of nutrient levels and phytoplankton production on the Red Cedar River during June 1958. Bars denote mean phytoplankton units ($\text{AA} \times 10^3 \text{ cm}^{-2} \text{ day}^{-1}$).



Comparison of nutrient levels and phytoplankton production on the Red Cedar River during June 1958. Bars denote mean phytoplankton units ($\text{AA} \times 10^3 \text{ cm}^{-2} \text{ day}^{-1}$).

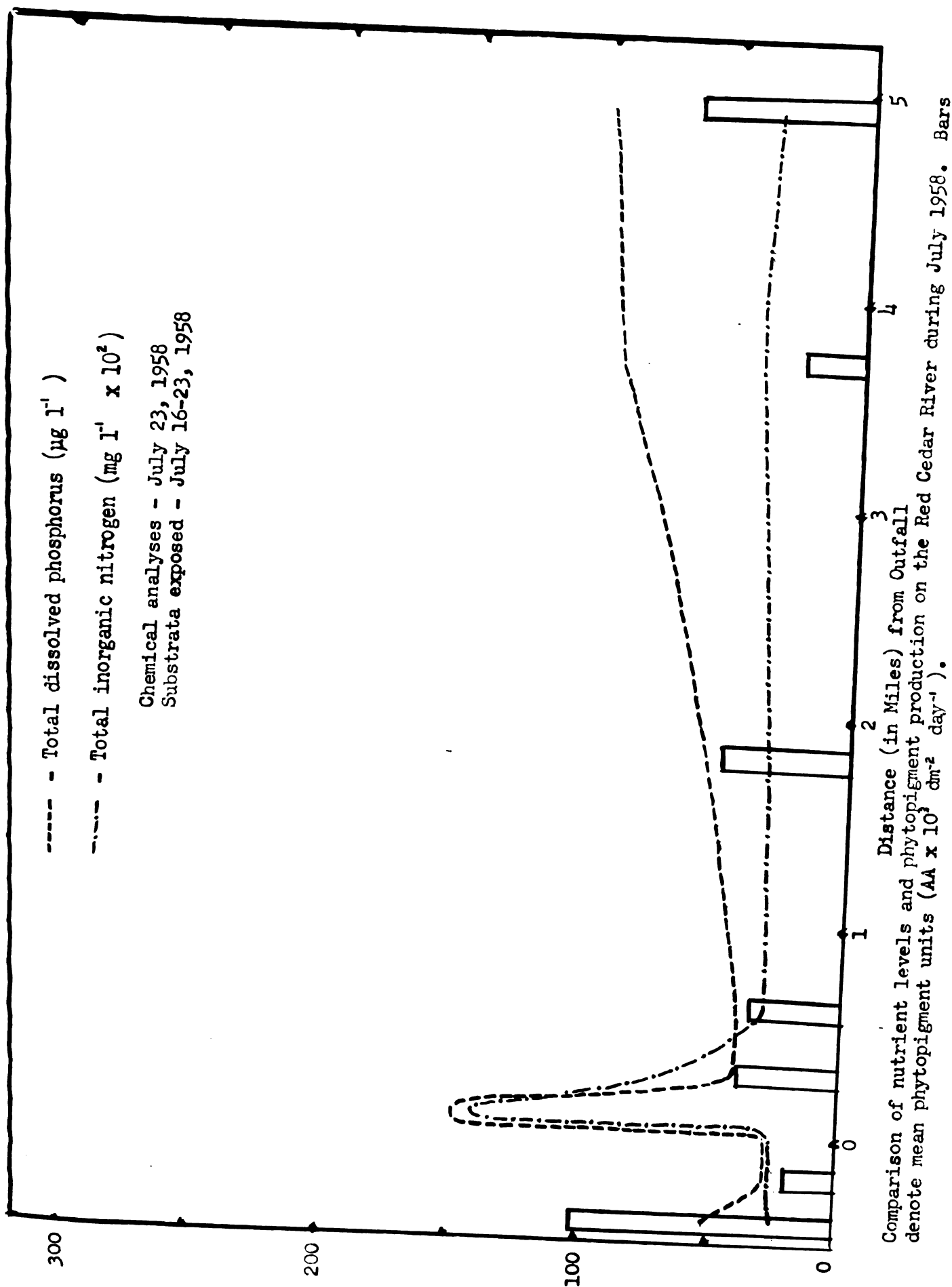
Substrata exposed June 26 to July 10, 1958
(4 foot rise in water level)

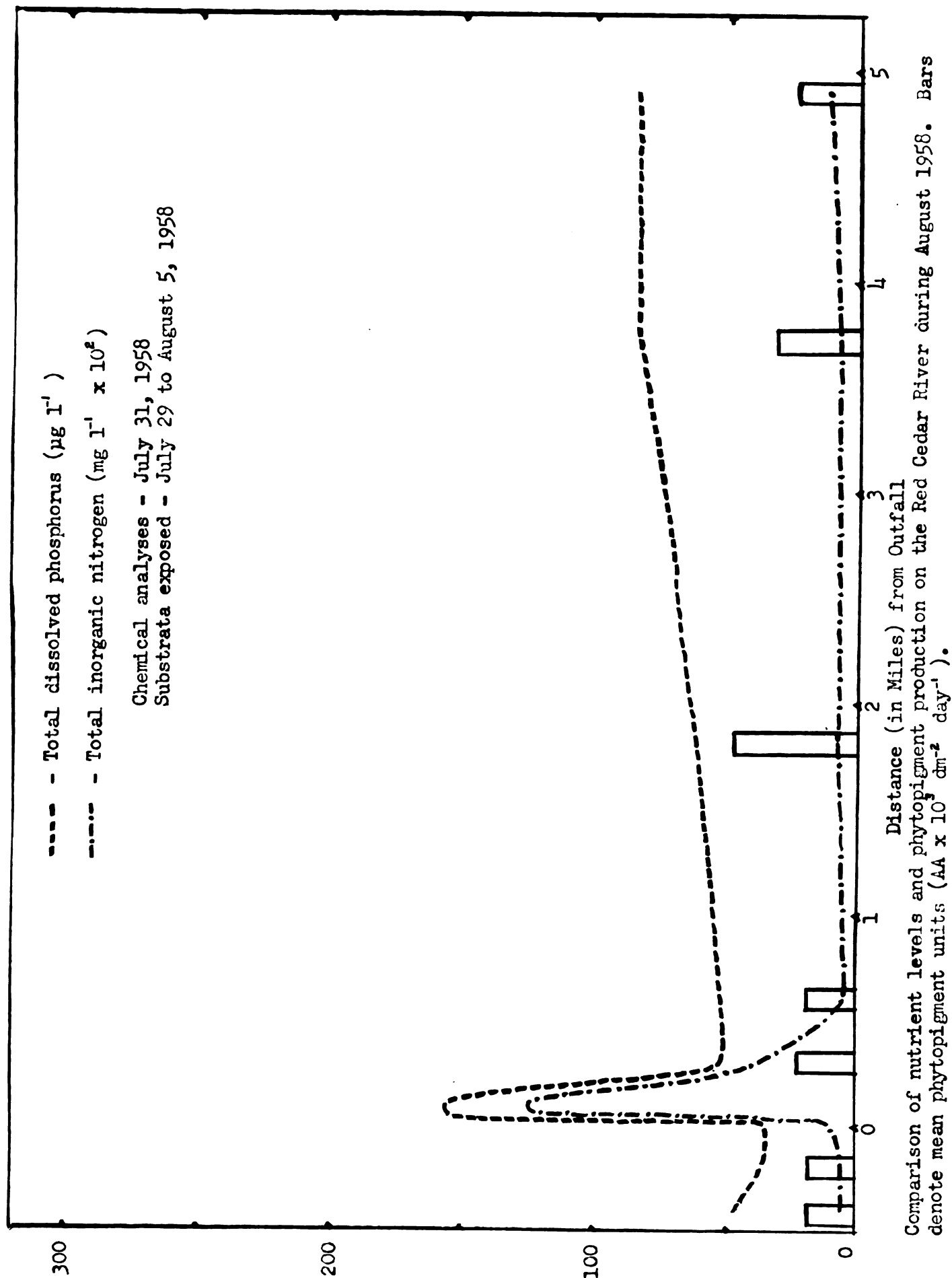


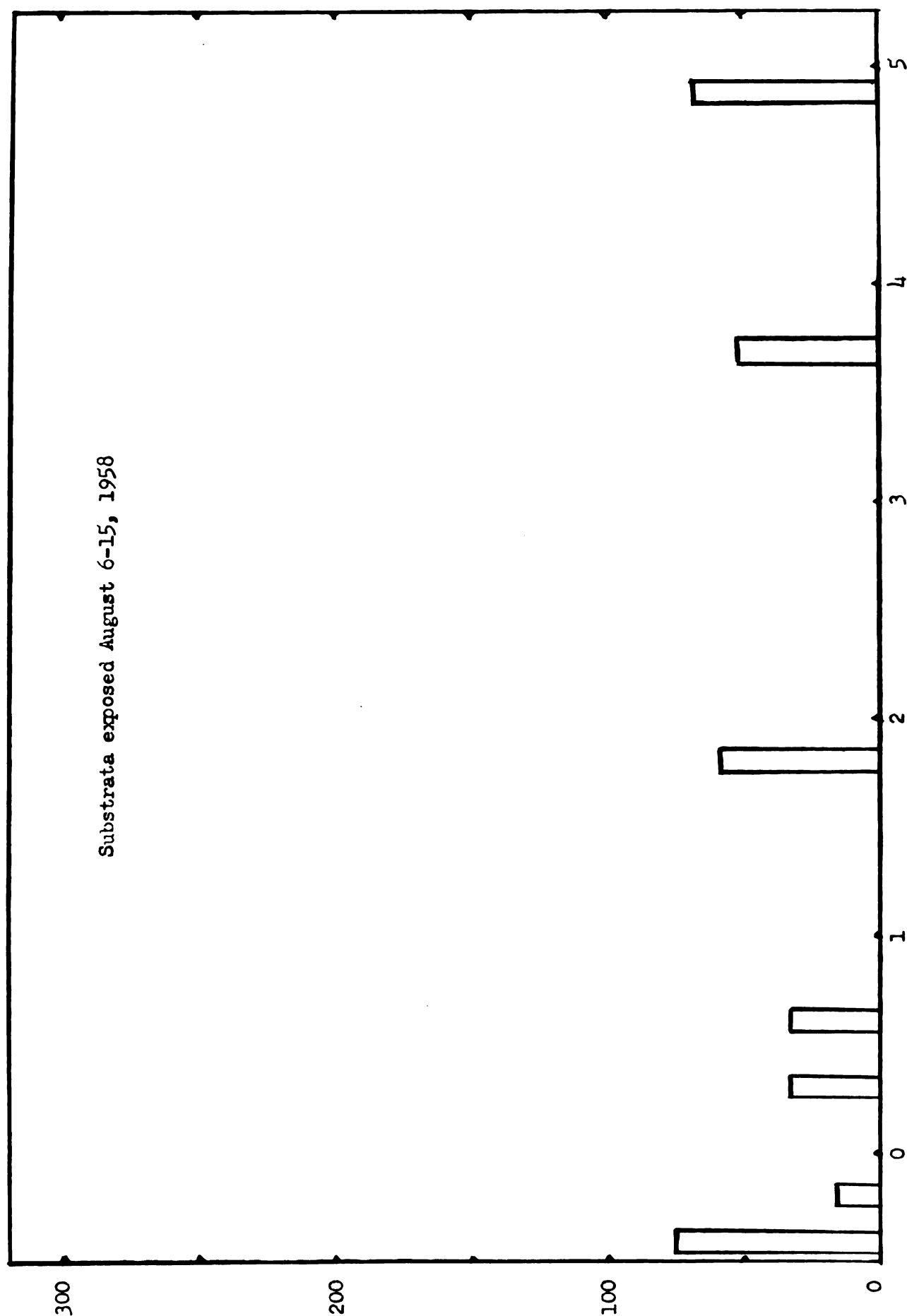
Phytoplankton production on the Red Cedar River during July 1958. Bars denote mean phytoplankton units ($10^3 \text{ dm}^{-2} \text{ day}^{-1}$).

----- - Total dissolved phosphorus ($\mu\text{g l}^{-1}$)
 - - - - - Total inorganic nitrogen ($\text{mg l}^{-1} \times 10^2$)

Chemical analyses - July 23, 1958
 Substrata exposed - July 16-23, 1958







Phytoplankton production on the Red Cedar River during August 1958. Bars denote mean phytoplankton units (AA x 10³ dm⁻² day⁻¹).

APPENDIX D

Nutrient levels of the Red Cedar River at Station -0.4

Date	Phosphorus ($\mu\text{g l}^{-1}$)			Nitrogen (mg l^{-1})		
	Dissolved	Sestonic	Total	Ammonia	$\text{NO}_2 + \text{NO}_3$	Total
July 5, 1957			109	0.0	1.86	1.86
27	44	73	117	0.0		
August 6	28	60	88	0.0		
12	41	65	106	0.0	0.28	0.28
20	51	62	113	0.0		
September 13	46	33	79	0.0	0.31	0.31
October 3	44	44	88	0.09		
31	38	17	55	0.20	0.41	0.61
November 19	28	15	43	0.09	0.68	0.72
December 5	29	27	56	0.11	1.23	1.34
January 14, 1958	24	29	53	0.14	0.90	1.04
February 6	31	36	67	0.17	0.82	0.99
March 11	22	21	43	0.06	1.53	1.59
26	18	24	42	0.0	1.06	1.06
April 8	15	38	53	0.09	1.04	1.13
15	13	34	47	0.0	0.86	0.86
22	16	64	80	0.0	0.73	0.73
30	13	55	68	0.0	0.30	0.30
May 12	19	37	56	0.0	0.11	0.11
15	15	41	56	0.0	0.21	0.21
27	23	55	78	0.0	0.0	0.0
June 2	37	74	111			0.21
10				0.33	0.26	0.59
19	35	42	77	0.14	0.25	0.39
July 1	32	65	97	0.0	0.11	0.11
5	38	44	82			2.80
7	44	34	78			1.22
9	26	46	72			1.20
23	53	31	84	0.0	0.26	0.26
31	47	60	107	0.0	0.06	0.06

Nutrient levels of the Red Cedar River at Station -0.2

Date	Phosphorus ($\mu\text{g l}^{-1}$)			Nitrogen (mg l^{-1})		
	Dissolved	Sestonic	Total	Ammonia	$\text{NO}_2 + \text{NO}_3$	Total
July 5, 1957			230	0.0	4.12	4.12
27	33	44	77	0.0		
August 6	26	52	78	0.0		
12	41	54	95	0.0	0.21	0.21
20	47	49	96	0.0		
September 13	41	30	71	0.0	0.41	0.41
October 3	44	21	64	0.11		
31	34	8	42	0.22	0.50	0.72
November 19	27	28	55	0.10	0.96	1.06
December 5	24	28	52	0.11	1.41	1.52
January 14, 1958	19	33	52	0.14	1.27	1.41
February 6	23	33	56	0.16	0.72	0.88
March 11	19	20	39	0.04	1.72	1.76
26	18	21	39	0.0	1.00	1.00
April 8	15	41	56	0.06	1.09	1.15
15	13	29	42	0.0	0.86	0.86
22	14	66	80	0.0	0.59	0.59
30	14	48	62	0.0	0.23	0.23
May 12	17	37	54	0.0	0.11	0.11
15	17	37	54	0.0		
27	22	50	72		0.0	0.0
June 2	29	69	98			0.29
10						
19	30	42	72	0.18	0.24	0.42
July 1	25	63	88	0.0	0.11	0.11
5	44	86	130			2.70
7	40	48	88			1.73
9	28	62	90			1.40
23	27	47	74	0.0	0.29	0.29
31	37	60	97	0.0	0.06	0.06

Nutrient levels of the Red Cedar River at Station #0.1

Date	Phosphorus ($\mu\text{g l}^{-1}$)			Nitrogen (mg l^{-1})		
	Dissolved	Sestonic	Total	Ammonia	$\text{NO}_2 + \text{NO}_3$	Total
July 5, 1957			230	0.0	4.12	4.12
27	52	63	115	0.31		
August 6	99	103	202	0.37		
12	208	59	267	0.70	0.18	0.88
20	248	90	338	0.47		
September 13	240	88	328	0.51	0.31	0.82
October 3	85	117	202	0.32		
31	98	69	167	0.53	0.46	0.99
November 19	75	22	97	0.32	1.30	1.62
December 5	133	62	195	0.60	1.25	1.85
January 14, 1958	44	65	109	0.28	1.22	1.50
February 6	126	90	216	0.65	0.82	1.47
March 11	59	43	102	0.19	1.80	1.99
26	88	54	142	0.31	1.00	1.31
April 8	150	110	260	0.57	1.20	1.77
15	93	63	156	0.25	0.86	1.11
22	76	154	230	0.37	0.64	1.01
30	148	124	272	0.50	0.28	0.78
May 12	252	248	500	0.69	0.12	0.81
15	82	26	108	0.32	0.24	0.56
27	240	152	392	0.78	0.30	1.08
June 2						
10						
19	162	48	210	0.82	0.24	1.06
July 1	222	158	380	1.09	0.21	1.31
5						
7						
9						
23	148	72	220	1.20	0.21	1.41
31	156	144	300	1.20	0.05	1.25

Nutrient levels of the Red Cedar River at Station +0.3

Date	Phosphorus ($\mu\text{g l}^{-1}$)			Nitrogen (mg l^{-1})		
	Dissolved	Sestonic	Total	Ammonia	$\text{NO}_2 + \text{NO}_3$	Total
October 3, 1957	74	42	116	0.27		
31	64	33	97	0.17	0.46	0.63
November 19	44	23	67	0.16	0.54	0.70
December 5	42	36	78	0.20	1.51	1.71
January 14, 1958	35	43	78	0.22	1.39	1.61
February 6	46	39	85	0.23	0.96	1.16
March 11	26	26	52	0.07	2.00	2.07
26	31	27	58	0.05	1.13	1.18
April 8	25	49	74	0.14	1.13	1.27
15	30	33	63	0.06	0.86	0.92
22	35	72	107	0.10	0.75	0.85
30	44	60	104	0.04	0.36	0.40
May 12	87	57	144	0.15	0.13	0.28
15	53	58	111	0.16	0.12	0.28
27	57	61	118	0.05	0.16	0.31
June 2						
10						
19	46	50	96	0.25	0.24	0.49
July 1	45	74	119	0.0	0.22	0.22
5						
7						
9						
23	43	60	103	0.43	0.24	0.67
31	51	60	111	0.35	0.03	0.38

Nutrient levels of the Red Cedar River at Station +0.6

Date	Phosphorus ($\mu\text{g l}^{-1}$)			Nitrogen (mg l^{-1})		
	Dissolved	Sestonic	Total	Ammonia	$\text{NO}_2 + \text{NO}_3$	Total
July 5, 1957						
29	46	57	103	0.0		
August 6	36	60	96	0.0		
12	46	56	102	0.0	0.14	0.14
20	54	59	113	0.0		
September 13	74	47	121	0.11	0.31	0.42
October 3	50	35	85	0.14		
31	38	24	62	0.16	0.97	1.13
November 19						
December 5	31	29	60	0.15	1.67	1.82
January 14, 1958	30	26	56	0.19	1.27	1.46
February 6	33	22	65	0.17	0.82	0.99
March 11						
26	22	22	44	0.0	1.00	1.00
April 8	26	38	64	0.10	1.20	1.30
15	17	33	50	0.0	0.49	0.49
22	15	70	85	0.0	0.61	0.61
30	23	48	71	0.0	0.46	0.46
May 12	84	60	144	0.08	0.14	0.22
15	26	45	71	0.0	0.12	0.12
27	41	57	98	0.0	0.08	0.08
June 2						
10						
19	42	52	94	0.16	0.10	0.26
July 1	37	75	112	0.0	0.12	0.12
5						
7						
9						
23	42	58	100	0.06	0.25	0.31
31	52	66	118	0.0	0.06	0.06

Nutrient levels of the Red Cedar River at Station +1.3

Date		Phosphorus ($\mu\text{g l}^{-1}$)			Nitrogen (mg l^{-1})		
		Dissolved	Sestonic	Total	Ammonia	$\text{NO}_2 + \text{NO}_3$	Total
May	12, 1958	26	46	72	0.0	0.10	0.10
	15	30	44	74	0.0	0.12	0.12
	27	48	63	111	0.0	0.11	0.11
June	2						
	10						
	19	44	67	111	0.15	0.11	0.26

Nutrient levels of the Red Cedar River at Station +1.8

Date	Phosphorus ($\mu\text{g l}^{-1}$)			Nitrogen (mg l^{-1})		
	Dissolved	Sestonic	Total	Ammonia	$\text{NO}_2 + \text{NO}_3$	Total
July 5, 1957			320	0.0	2.13	2.13
27	33	44	77	0.0		
August 6	38	48	86	0.0		
12	57	44	101	0.0	0.14	0.14
20	64	49	113	0.0		
September 13	51	46	97	0.0	0.31	0.31
October 3	73	42	115	0.14		
31	34	18	52	0.10	0.64	0.74
November 19	29	33	62	0.09	0.90	0.99
December 5	34	26	60	0.12	1.33	1.45
January 14, 1958	24	33	57	0.14	1.07	1.21
February 6	31	34	65	0.16	0.96	1.12
March 11	20	22	42	0.04	1.92	1.96
26	20	23	43	0.0	1.00	1.00
April 8	15	39	54	0.06	1.24	1.30
15	17	33	50	0.0	0.59	0.59
22	18	60	78	0.0	0.51	0.51
30	21	41	62	0.0	0.40	0.40
May 12	37	34	71	0.0	0.10	0.10
15	37	39	76	0.0	0.08	0.08
27	59	43	102	0.0	0.06	0.06
June 2	51	65	116			0.28
10				0.29	0.26	0.55
19	50	61	111	0.16	0.31	0.37
July 1	60	79	139	0.0	0.17	0.17
5	38	110	148			2.80
7	46	78	124			1.60
9	45	72	117			1.40
23	57	61	118	0.06	0.25	0.31
31	62	60	122	0.0	0.08	0.08

Nutrient levels of the Red Cedar River at Station #3.7

Date	Phosphorus ($\mu\text{g l}^{-1}$)			Nitrogen (mg l^{-1})		
	Dissolved	Sestonic	Total	Ammonia	$\text{NO}_2 + \text{NO}_3$	Total
July 5, 1957						
27	41	35	76	0.0		
August 6	74	28	102			
12	104	23	127	0.0	0.25	0.25
20	114	26	140	0.0		
September 13	90	19	109	0.0	0.51	0.51
October 3	73	24	97	0.06		
31	46	14	60	0.11	0.65	0.76
November 19	32	27	59	0.09	0.60	0.69
December 5	39	26	65	0.12	1.33	1.45
January 14, 1958	38	26	64	0.16	1.00	1.16
February 6	40	28	68	0.20	1.27	1.47
March 11	22	26	48	0.04	1.62	1.66
26	18	25	43	0.0	0.67	0.67
April 8	17	39	56	0.0	1.24	1.24
15	17	33	50	0.0	0.64	0.64
22	27	49	76	0.0	0.42	0.42
30	28	34	62	0.0	0.40	0.40
May 12	37	38	75	0.0	0.10	0.10
15	48	37	85		0.16	0.16
27	68	34	102	0.0	0.08	0.08
June 2						
10						
19	65	28	93	0.11	0.30	0.41
July 1	91	44	135	0.0	0.24	0.24
5						
7						
9						
23	93	33	126	0.0	0.39	0.39
31	84	37	121	0.0	0.13	0.13

Nutrient levels of the Red Cedar River at Station #4.9

Date	Phosphorus ($\mu\text{g l}^{-1}$)			Nitrogen (mg l^{-1})		
	Dissolved	Sestonic	Total	Ammonia	$\text{NO}_2 + \text{NO}_3$	Total
January 14, 1958	35	30	65	0.17	1.50	1.67
February 6	35	34	69	0.20	2.04	2.44
March 11						
26	19	20	39	0.0	0.89	0.89
April 8	20	41	61	0.0	1.04	1.04
15	20	36	56	0.0	0.57	0.57
22	27	57	84	0.0	0.37	0.37
30	26	33	59	0.0	0.40	0.40
May 12	37	38	75	0.0	0.03	0.03
15	50	35	85	0.0	0.16	0.16
27	71	31	102	0.0	0.07	0.07
June 2						
10				0.29	0.35	0.64
19	65	39	104	0.11	0.33	0.44
July 1	97	47	144	0.0	0.27	0.27
5						
7						
9						
23	100	40	140	0.0	0.35	0.35
31	85	39	124	0.0	0.13	0.13

APPENDIX E

Physical and chemical characteristics of the Red Cedar River
as determined at Station #3.7.

		Water Temperature (°C.)	Conductivity ($\times 10^{-6}$ ohms $^{-1}$ cm $^{-1}$)	Alkalinity (mg l $^{-1}$)	pH	Turbidity (Units)
July	5, 1957	20.0		192	7.9	352
	27	22.0	634	296	7.9	68
August	6	20.0	606	286	7.9	38
	12	21.0	588	276	8.1	17
	20	21.0	519	266	8.2	26
September	13	20.0	562	260	8.1	19
October	3	11.5	460	270	8.0	12
	31	6.5	466	270	7.8	14
November	19	5.0	540	228	7.7	15
December	5	0.5	621	272	8.0	22
January	14, 1958	0.0	573	276	7.9	23
February	6	0.0	610	284	8.0	15
March	11	1.0	515	220	8.1	14
	26	4.0	528	244	8.4	13
April	8	7.0	510	226	8.1	25
	22	14.5		266	8.3	22

ROOM USE ONLY

ROOM USE ONLY

~~MAR 31 1961~~

~~SEP 27 1962~~

~~OCT 11 1962~~

~~MAR 15 1963~~

~~APR 2 1963~~

~~FEB 19 1964~~

~~APR 23 1964~~

~~JUL 15 1964~~

~~JUL 22 1964~~

~~NOV 25 1964~~

~~DEC 7 1964~~

~~FEB 1 1965~~

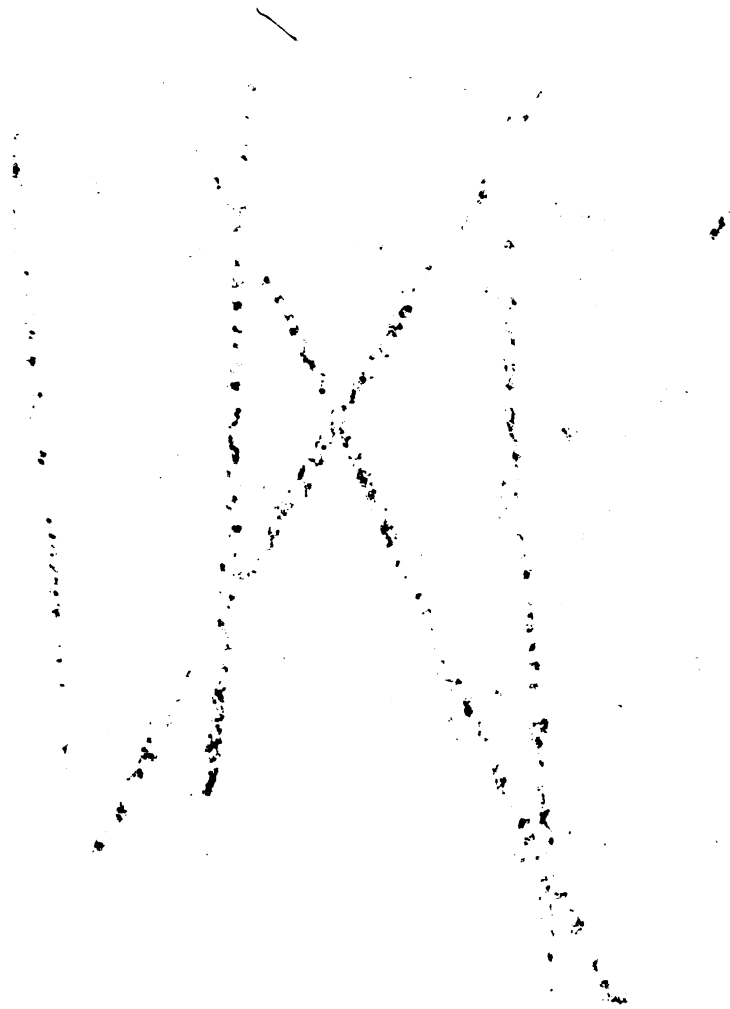
~~MAR 1 1965~~

~~MAR 15 1965~~

APR 12 1965

MAY 8 1965

APR 3 1966



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