January 1971

Detergent and Non-Detergent Phosphorus in Sewage

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to

The Procter and Gamble Company
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Introduction

Quantitative data on nutrient levels in sewage from homes which are using heavy-duty detergents and which have been restricted from using such detergents is necessary before rational decisions on the removal of phosphorus from detergent can be made. Furthermore, the effect of various standard sewage treatment schemes on phosphorus levels should be evaluated to determine the relative cost of such treatment. Then the cost (to society, the environment, and to the taxpayer) of phosphorus removal from detergents and/or from sewage effluents can be estimated and such data utilized to make the appropriate policy decisions.

The report herein presented is concerned principally with developing basic data that will provide information as to the changes that occur in concentration, biostimulation, and phosphorus removal efficiencies with and without heavy-duty detergents.

Experimental Design

The experimental design was based on the measurement of differences in nutrient concentrations in sewage caused by changes in habits in a selected population of homeowners. Also the measurement of differences in nutrient concentrations caused by typical methods

of treating such sewages was determined. The differences were measured by chemical analysis, and batch and chemostat (data to be reported elsewhere) bioassay techniques.

The selected population was composed of a middle to upper middle class suburban community that was relatively isolated from the rest of the City of Logan. The 61 homes (59 occupied) in the study area were located at the end of the sewer system which made it possible to collect samples from the area without the influence of other areas. The area is also located at an elevation that eliminates groundwater problems associated with infiltration, and the sewers are constructed with gasket type joints and are less than ten years old. With this location and system, it was felt that the samples would provide unbiased data.

The study schedule was as follows:

By March 31.	Planning and mobilization of the study were completed.
April 7, Wednesday.	The baseline sample was collected on this date without informing the study neighborhood of the project.
April 8-May 7.	Nutrient and bioassay analyses of the baseline sample were performed.
April 16-26.	An explanation of the project to the homeowners in the study neighborhood was conducted with

group meetings and individual contacts.

April 26-28. Period when use of dishwashers and clothes-washers was prohibited.

April 28, Wednesday. The collection of the test sample was completed.

April 29-June 5. Analysis of the test sample was completed with the exception of the chemostat analyses which were delayed. These results will be submitted as a separate report.

Reservoir Water

Most of the bioassays were performed on dilutions of the treated effluents in reservoir water collected from First Dam Reservoir on the Logan River (Figure 1). The Logan River begins in the northerly portion of the Wasatch Mountains in an area where some of the peaks are slightly higher than 10,000 feet. The drainage basin itself is almost completely within the Cache National Forest and is subjected to some grazing, winter and summer recreation, summer houses and campsites, and hunting and fishing. No communities discharge into the Logan River upstream of Logan City. At the time of sampling (April-May 1971) human activity in the drainage basin was minimal because the spring thaw was beginning.

The First Dam Reservoir itself is upstream of Logan City and near the Utah Water Research Laboratory where the research was

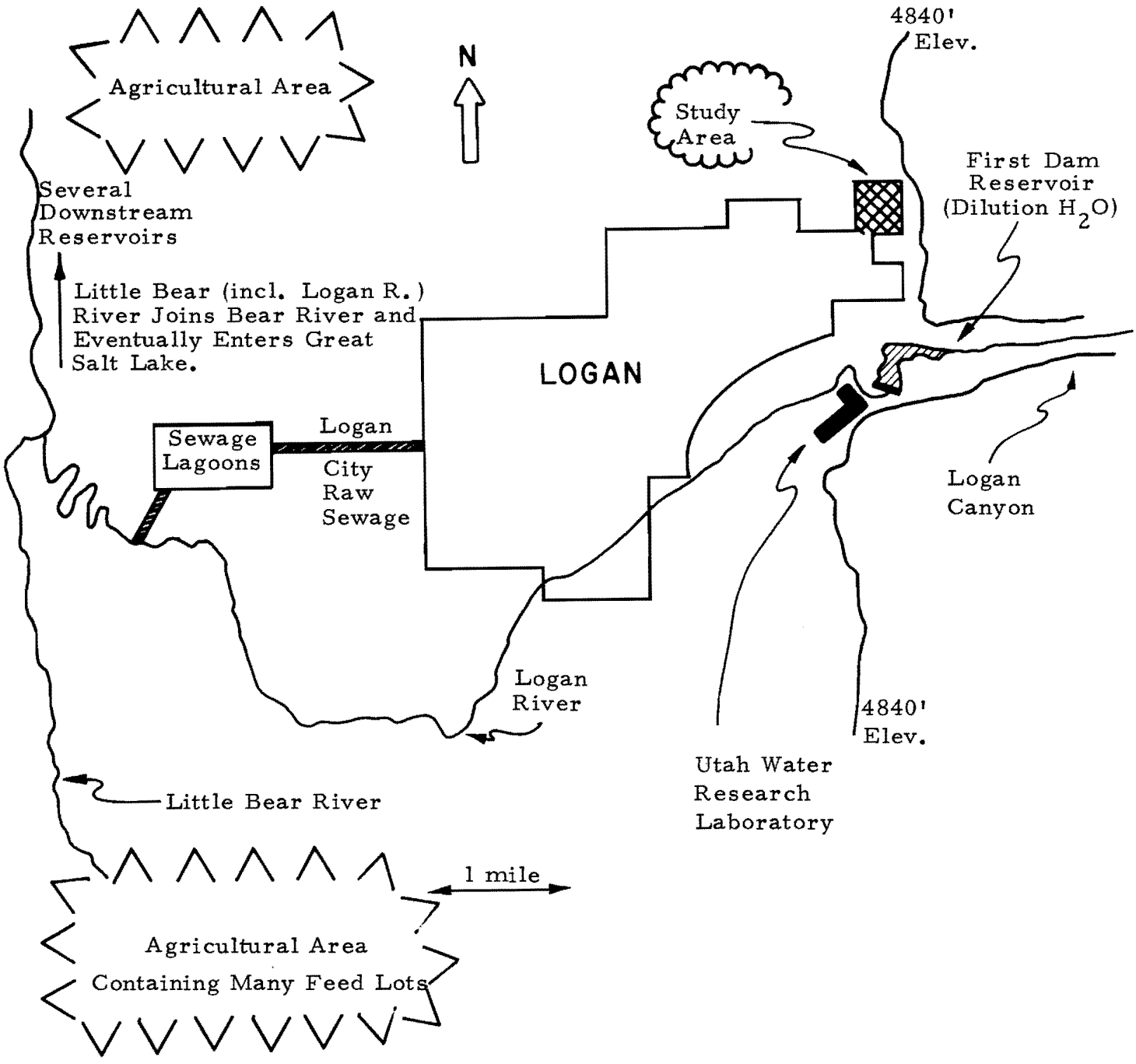

Figure 1. Sketch of Logan River System showing Reservoir, Sewage Lagoons. Input of river water from agricultural area, and relation to City of Logan.

accomplished. Sewage from Logan City is discharged into sewage lagoons before being released into the Logan River in the approximate center of Cache Valley. The Logan River joins the Little Bear River and eventually the Bear River which terminates in the Great Salt Lake. In Cache Valley most nutrient input probably originates from agriculture.

Study Population

A map of the neighborhood studied is shown in Figure 2 and consists of 61 houses, 59 of which were occupied. All percentages reported herein were calculated based on the 59 residences.

Contact with Homeowners

Cooperation of the homeowners was obtained by the following sequence of contacts:

1.) Hand delivery of informational letters along with the questionnaire, affidavit, instruction, and billing sheet were delivered on Sunday, April 18, 1971. (See Appendix A-1.)

2.) A meeting was held on Thursday, April 22, 1971 at 7:30 PM to show slides, discuss the project, answer questions, and obtain the names of people participating in the study. (See Appendix A-2 for signup sheet.) Paper plates and cups were distributed to participants present at the meeting.

3.) The cooperation of the remaining people was solicited on

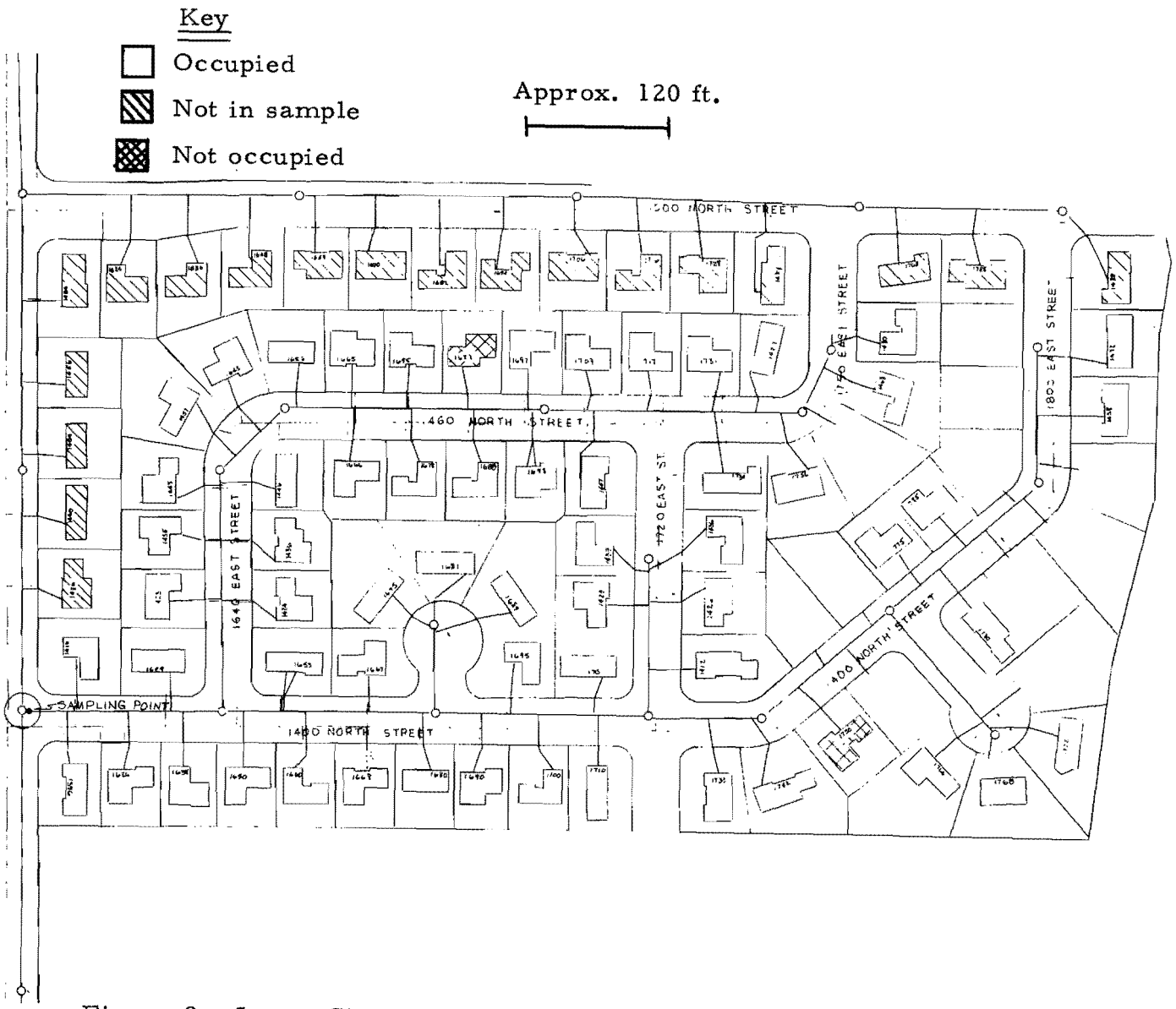

Figure 2. Logan City Map: Detail of Study Neighborhood.

Friday and Saturday, April 23 and 24, 1971, and paper plates and cups were distributed.

4.) Bottles of soap (12 bottles of Joy, 23 bottles of Dove, 8 bottles of Ivory, 16 bottles of Lux; all 22 oz. bottles), a reminder sheet (Appendix A-3), tape (labeled "Phosphate Study"), and postage stamps for returning questionnaires, etc., were distributed on Sunday, April 25, 1971.

5.) Attended a coffee hour from 9-11 AM Monday, April 26, 1971, and showed slides and answered questions of neighborhood housewives to insure the cooperation of the housewives.

6.) The neighborhood refrained from using dishwashers and clotheswashers during Monday to 01:00 AM Thursday (April 26-29, 1971). Sampling was from 05:00 AM Wednesday (April 28) to 01:00 AM Thursday (April 29).

7.) The affidavits and questionnaires were collected on Thursday and Friday (April 29 and 30).

8.) Mailed congratulatory letter to residents on Wednesday, May 26, 1971 (see Appendix A-4). Note that an error in the listed phosphorus concentrations and percent decrease in phosphorus concentration occurred. This was due to a miscalculation which was later corrected when results were checked.

Reactions of Homeowners

As subjectively judged by personal contact, the residents exhibited one of three attitudes (14 percent no assessment available): 59 percent enthusiastic cooperation ("what more can I do?"), 20 percent cooperation ("but don't bother me anymore"), and 7 percent unwilling cooperation. No one refused to cooperate although some residents thought we would be lucky to get 80 percent cooperation. We received 59 signed affidavits and completed questionnaires. The second attitude may have resulted from the number of times we knocked on their doors, but if we had not made 100 percent contact, we most probably would have had less than 100 percent cooperation. With regard to specific individuals, one household was never contacted by us, but told one of the ladies helping us (Mrs. P. Koenig) that she would cooperate but did not want any free plates, cups, or soap and did not want to be bothered. She later mailed in an affidavit and questionnaire. Another lady signed an affidavit and then noted that she had used her clotheswasher twice on the sampling day (Wednesday, April 28, around 1 PM). It is possible that we missed the effect of that detergent, and also because the flow was small during the afternoon very little effect of that detergent use on phosphate concentrations would be expected in the final composite (see Sampling Methods below). It is possible that others did the same but did not admit it. This will be discussed below.

After the sampling had been completed, four people said they would not be willing to participate further in the study and six were not sure. This reaction was not unexpected because these were probably people who objected to the upsetting of their routine.

Only three persons submitted bills. Because women with several small children still in diapers had somewhat of a hardship, we offered to buy disposable diapers for them. Thus, the three bills totaled $7.39 of which $6.45 was for disposable diapers.

Use Statistics of Selected Population

In the 59 residences there were 252 individuals, of which 125 were under 18 years of age (Table 1). There were 49 dishwashers (0.83/house) and 60 clotheswashers (1.02/house) in the 59 homes indicating a rather high use of detergent. Estimates of detergent use were considered relatively poor and are not included. Garbage grinders (0.95/house), bath-shower facilities (2.14/house), and toilets (2.39/house) indicate high water use. Only 19 homes had water softeners, 8 of the 19 solely for hot water. Thus, less detergent would be required in approximately 1/3 of the homes. In general it would be expected that higher phosphorus concentrations would be observed in the test area. However, some women volunteered that they were using low-P detergents (Sears). This would lower the phosphorus level in the baseline sample, but because they were not allowed to use this or any detergents in the

Table 1. Water use facilities description.

Facilities	Total	Number per occupied house
Inhabitants (under 18)	252 (125)	4.27 (2.12)
Occupied Homes (total homes[a])	59 (61[a])	-
Water Softeners	19	0.322
Heat Hot Water Only	8	-
Water Use Facilities		
Toilets	141	2.39
Dishwashers	49	0.83
Clotheswashers	60	1.02
Garbage Grinders	56	0.95
Bath or Shower	126	2.14
Sinks	222	3.76
Bathroom	150	2.54
Other	72	1.22

[a] 2 unoccupied houses were not included in any calculations.

Table 2. Specific questions concerning use of materials which would possibly affect results (46 answered; 13 did not).

	Yes	No	Unsure
1. Did you use paper plates and cups? (Amount not answered.)	34	11	1
2. Did you use any kinds of soaps or cleansers? (Kinds not answered.)	14	31	1
3. Did you change habits from normal in any way? (How not answered.)	16	28	2
4. Did you use disposable diapers in exchange for washables? (Amounts not answered.)	8	36	2

study, it would not affect the test sample.

After sampling, estimates were made of other factors which might have affected the results by direct person-to-person questioning. Answers were quite variable and indefinite and only 78 percent of the residents provided answers so little can be determined (Table 2). Questions about type and quantity of factors that might have affected the study were considered unanswered because people were so unsure. Roughly three quarters of the people used the paper plates and cups while about 30 percent used soaps and cleansers in activities other than involving dishwashers or clotheswashers. About 40 percent reported an effect on their habits. Eight persons reported using disposable diapers in exchange for washables.

The effect of these factors on the composition of the sewage would be variable, some diluting and some increasing nutrient concentrations while other factors would have an effect on total flow. It will be assumed that the factors would be somewhat self-canceling, and that their effect would be minimal.

Procedures and Methods

Sewage Sampling

Time and place

The location of the study population was based on the following criteria: 1) low infiltration, 2) easily isolated, and 3) relatively

homogeneous suburban population. The study area is located at the end of a sewer line and composed of 88, relatively new, single-family residences of which 59 residences (61 houses) were isolated. The area is located such that it is above the major groundwater areas (minimizing infiltration), and the sewer lines have rubber gasket seals, and little if any surface drainage enters the system. Storm water is carried off by surface drainage only and does not enter the sewage system in the test neighborhood.

Both the baseline and test samples were collected on a Wednesday (05:00 AM Wednesday to 01:00 AM Thursday) while school was in session with a three week interval separating sampling (April 7 and April 28). The weather was only slightly warmer on April 28; rain showers occurred on both sampling days.

Method of sampling

An eight-inch diameter, 90^{o} angle, V-notch weir was placed in the sewer line which drains the neighborhood (calibration and conversion charts in Appendix B-1). The weir was cleaned and the head gage on the weir was read just prior to sampling. A sampling scoop was constructed from the bottom half of a one-gallon polyethylene container attached to a wooden handle. Samples were collected at one-half hour intervals and the moment of sampling was defined by a bell on a timer. When the bell rang, the scoop was lowered into the pipe downstream of the weir until all of the flow entered the sampler. This sample

was then poured into a labeled sample bottle. Usually several scoops were required to fill one sample bottle. Larger volumes were collected for higher flows to insure adequate sample for compositing. The temperature was read after sampling from a thermometer in the waste stream. Samples were transported to the lab (at 2-3 hour intervals) to be stored in the refrigerator (5^{o} C).

Compositing of Sample

Flow rates were calculated and the total volume of the highest flow rate sample was used in the composite sample. The volume required from the other samplings was calculated on the basis of the ratio of the sample flow to the highest flow obtained. Each sample was placed in a Waring blender on the highest setting for 15 seconds before taking the appropriate volume for the composite sample. Thus, the raw sewage sample was composed of 40 subsamples (20 hours of sampling at half-hour intervals).

Sewage Treatment

The composited raw sewage sample was placed in a large (50 gallon) polyethylene tank and mixed well using a large paddle. Subsamples were collected for analysis. Activated sludge seed was added and aeration begun. The activated sludge seed was started with a raw sewage sample plus dogfood and nutrients as feed. One week prior to taking the baseline sample, sewage was collected from the test neighborhood

and used to acclimate the seed. Then after settling the seed was added to the raw sewage sample. After settling of the activated sludge and siphoning off of the secondary effluent, the activated sludge was placed in the refrigerator (5° C) until one week prior to collecting the test sample. At that time the sludge was removed and a raw sewage sample was added and aeration begun to prepare it for adding to the raw test sewage sample.

The secondary treatment was not typical because of its batch operation and the desire to minimize the chemical effect of adding the seed to the actual sample. Thus, the mixed liquor volatile suspended solids (236 mg/l) was considerably lower than typical (1500-2500 mg/l) and the residence time was considerably longer. Secondary treatment was probably more similar to extended aeration than conventional activated sludge. Secondary treatment was considered terminated when removal of the estimated BOD was 80-90 percent. The secondary effluent was collected by siphoning off the clear supernatant remaining after 1 to 2 hours of settling of the mixed liquor. This supernatant was then well mixed, sampled, and the remainder passed to tertiary treatment.

Tertiary treatment was performed by adding 300 mg/l of alum ($Al_2(SO_4)_3$) to the secondary effluent. Three minutes of manual rapid mixing with a paddle was followed by thirty minutes of slow mixing with a mechanical stirrer. Then the supernatant was siphoned off after 30 minutes of settling, mixed well, and sampled. This process worked

well for the baseline sample, but "floc" did not form in the test sample. Various jar tests at various concentrations of coagulant were tried but did not yield usable information. Therefore, one hour of slow mixing was tried and the sample was allowed to settle overnight before drawing off the supernatant.

All samples were stored in the refrigerator (5^{o} C) in polyethylene bottles until analyzed.

Chemical Analyses

All six samples (raw, secondary, and tertiary of baseline and test samples) were analyzed using the methods described below:

Alkalinity[2] was measured on an unfiltered sample and the initial temperature and pH (Beckman Zeromatic) were recorded to enable calculation of inorganic carbon according to Saunders et al.[1]

Conductivity was measured in an unfiltered sample. The conductivity of a standard KCl solution was measured at the same time and temperature recorded so that corrections could be made.[2]

Carbon was measured on filtered (GF/C, Whatman) and unfiltered samples with both the total and inorganic channel of a Model 915 Beckman TOCA.[3] Samples from the bioassay flasks were analyzed similarly but using only the total channel.

Suspended Solids (SS) and Volatile Suspended Solids (VSS) were measured using Whatman GF/C filters (for both sewage and bioassay

samples) which had been prerinsed and ashed (435° C) for 20 minutes prior to filtration. VSS were determined as the ash weight (560° C) difference.[2]

Iron was measured using the phenanthroline technique[2] on both filtered (0.45 μ MP) and unfiltered samples.

Phosphorus was measured using a modification of the Murphy and Riley technique.[4] Analyses were performed on unfiltered, persulfate digested[5] samples (total P) and on filtered (0.45 μ MP) samples (orthophosphate-P).

Nitrogen was measured in its common inorganic forms (NH_4^+, NO_3^-, NO_2^-) and as total organic nitrogen. The NH_4^+ was measured colorimetrically on an unfiltered sample.[6] The NO_3^- and NO_2^- ions were measured colorimetrically[2,4] on filtered samples but the NO_3^- was first converted (by Cadmium Reduction[4]) to NO_2^- and 0.95 (NO_2^-) subtracted from the total to obtain NO_3^-:

$$NO_3^- = \text{Total } (NO_3^- + NO_2^-) - 0.95\ (NO_2^-).$$

Total organic nitrogen was measured on unfiltered samples according to Standard Methods.[2]

5-day Biological Oxygen Demand (BOD) was measured on 3 different dilutions of unfiltered samples. A 0.2 percent v/v seed (raw sewage sample) was added only to the tertiary effluent. Some difficulties were experienced with the tertiary effluent from the test sample. There seemed to be

considerable initial oxygen demand which could not be satisfied by aeration. Very low dilutions were therefore used but complete depletion was still obtained. It was assumed that less BOD was present than in the secondary sample.

Nutrient Analyses (N, P, Fe) were also performed for the First Dam Reservoir on the Logan River and on some of the diluted samples for bioassay as a check for accuracy and precision. Total P and/or orthophosphate-P were measured on selected samples of homogenized, raw sewage from the test series to check on detergent usage. Also the quantity of total P was measured in the gift liquid detergents and in the add back detergents.

Instruments used for colorimetric analysis were the Beckman Model B (1 cm and 5 cm cells) and the B&L Spectronic 20 (1/2" and 1" cells). Standard curves were prepared and results indicate good agreement between both instruments.

Bioassays: Algal Assay Procedures: Bottle Test

Methods were adapted from the October, 1970, PAAP.[7] Light was 400 foot candles (maximum range of exposure was ± 10 foot candles); temperature was maintained at 25 ± 1° C; flasks were 500 ml pyrex Erlenmeyer flasks, capped with 150 ml pyrex beakers, containing 250 ml of sample plus inoculum. The pH was monitored periodically, and ventilation with filtered, scrubbed air (see Appendix B-2) was begun

when the pH value was ≥ 8.3. Flasks were not shaken and were hand mixed only when sampling.

Measurements of biomass (particulate carbon, absorbance at 750 mμ, suspended solids) were made at the end of seven days and when growth had ceased (≥ 18 days). Particulate carbon was determined as the difference between the total carbon concentration of filtered (GF/C) and unfiltered samples. The unfiltered sample was sonicated for ~ 15 seconds prior to carbon analysis.

Both Selenastrum capricornutum and Anabaena flos-aquae (cultures provided by T. E. Maloney, EPA, Corvallis, Oregon) were used as inocula at initial concentrations of 1000 cells/ml and 50,000 cells/ml, respectively. Occasionally old cultures (> 15 days) of A. flos-aquae required scraping with a rubber policeman to loosen cells attached to the walls of the flasks.

Results

Characteristics of Sewage Samples

There was considerable variability in the pattern of sewage flow between the two sampling dates (Figure 3). Also the total flow differed, increasing by 22 percent from the baseline to the test sample (Table 3). This variation could have been caused by people being out of town on April 7 (all 59 homes were apparently occupied on April 28; no data available for April 7) or an influx of party-goers on April 28. At least

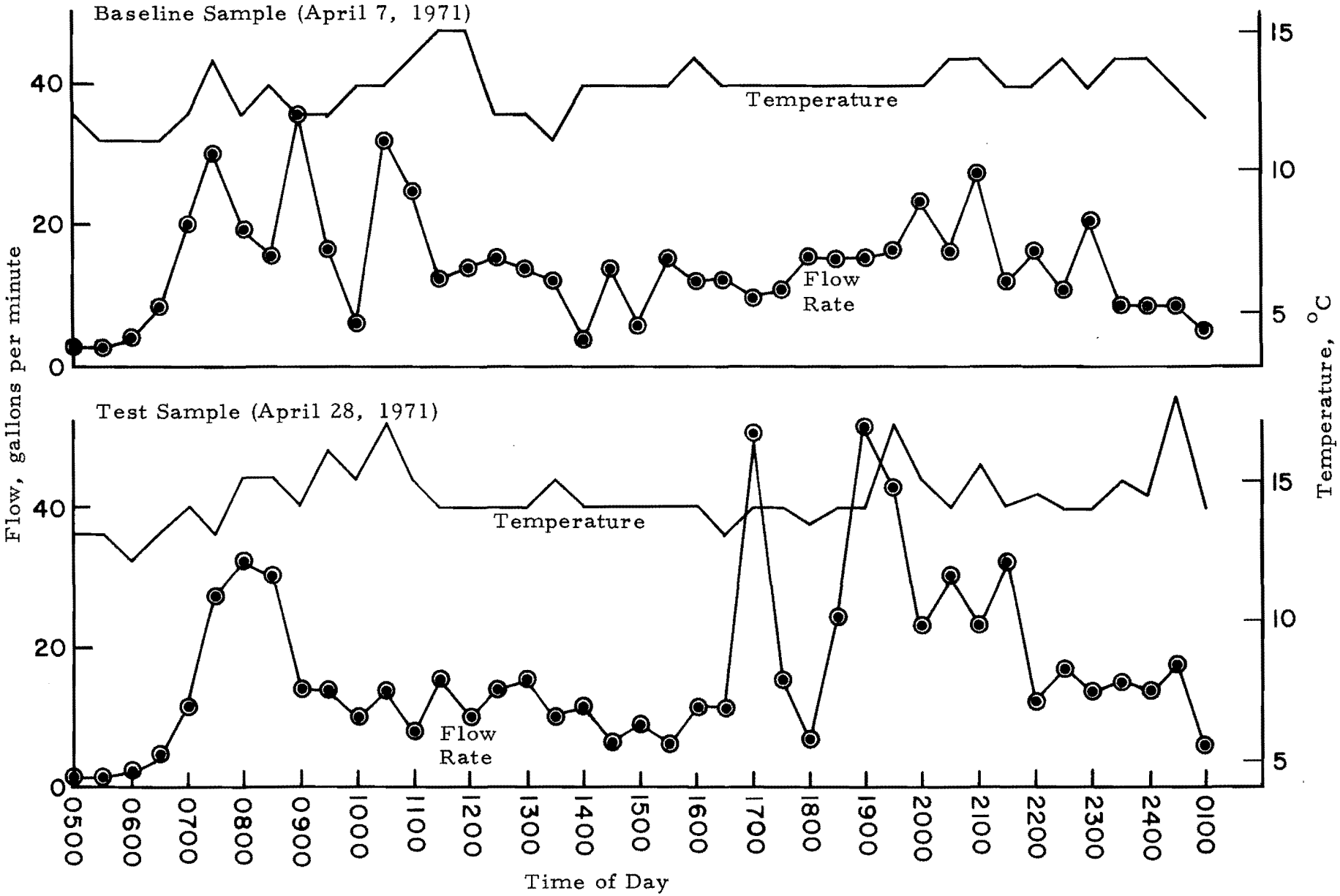

Figure 3. Sewage Flow and Temperature Variations for Two Different Sampling Dates.

Table 3. Characteristics of sewage samples.

	Samples	
	Baseline (April 7)	Test (April 28)
Total Flow, gals/day[a]	18,000	22,000
Mean Flow, gals/min[b]	15.0	18.3
Maximum Observed Flow, gals/min	36.0	51.5
Minimum Observed Flow, gals/min	2.8	1.6
Flow per capita, gals/day·person	71	88
Flow per household, gals/day·household	305	373

[a]Computed only for 20 hour period (0500 to 0100). Remaining 4 hours (0100 to 0500) estimated to affect final total by < 3%.

[b]Mean Flow is (Total Flow)/(20 hrs·60 mins/hr).

Table 4. Decrease in sewage phosphorus concentrations caused by restriction of detergent use and the effects of treatment.

	Concentration, mg Phosphorus/l		Percent Removal Total Phosphorus from Raw Sewage
	Orthophoshpate	Total Phosphorus	
Baseline Sample			
Raw Sewage	9.4	18.7	-
Secondary Effluent	12.8	17.0	9
Tertiary Effluent	0.041	0.65	97
Test Sample			
Raw Sewage	4.7	8.1	-
Secondary Effluent	4.2	6.5	20
Tertiary Effluent	0.033	0.25	97

one house had a dinner party for 20 on Wednesday (April 28) and this may explain the peak flows observed from 6:30 to 9:30 PM (Figure 3). In such a small population a temporary change in population or a change in habits could have a rather dramatic effect on flow rates. However, the flow rate change had little apparent effect on the nutrient study results as will be seen below.

The flow per capita is considerably less than estimated for municipalities as a whole (150 gal/day · person) but somewhat higher than for "typical" residential areas; a neighborhood having an average assessed valuation of $10,000 would produce 56 gal/day · person.[8]

The mean temperature of the sewage was slightly higher (about 13° C on April 7 and about 14° C on April 28) as would be expected as the weather continued to warm. Generally changes in sewage temperature coincided with changes in flow rate. Temperature increase would be associated with hot water use as with bathing and clothes and dishwashers. Visual observations of "detergent" suds in the sewage generally coincided with increases in sewage temperature for the baseline sample. For the test sample "detergent" suds were observed only at 10:30 in the morning and 9:00 in the evening. In contrast to the test sample, an adjacent sewage line draining a neighborhood which was not in the study showed obvious "detergent" suds at times of the day which matched the distribution of the baseline sample. Thus it was concluded that the participating householders were quite careful to follow the restrictions imposed upon

them. Chemical analyses reported below further substantiate this argument. This lends support to the argument that the increased flow of the test sample was caused by an influx of population to the neighborhood.

Chemical Analyses

Chemical analysis showed the considerable effect of restricting detergent use and of alum treatment upon phosphorus concentrations in sewage and treated sewage effluents (Table 4). Comparison of the baseline and test raw sewages showed that a 57 percent decrease in the total phosphorus occurred because of the restriction in dishwasher and clotheswasher use. The test sample itself was relatively, uniformly low in orthophosphate and total phosphate. Measurements of randomly selected grab samples indicated that the people in the neighborhood were conscientious about avoiding detergent use (Table 5). The one high value was the 5:00 PM total phosphorus measurement which was probably a result of high fecal content observed in that sample. Substitute detergents were handed out to the neighborhood and these would contribute little phosphorus to the sewage (Table 6).

Analyses of other chemical nutrients (Table 7) showed that organic carbon and BOD decreased about as expected with treatment while nitrogen (ammonia) remained approximately constant except for the reduction in organic nitrogen. Little nitrification took place as evidenced by the very low concentrations of nitrite plus nitrate. Iron was relatively low in the raw sewage but was still probably adequate for significant algal growth in tertiary effluents. Alkalinity, pH, and conductivity all varied as expected; the addition of alum for tertiary treatment reduced the alkalinity

Table 5. Concentrations of orthophosphate and total phosphorus in selected grab test samples collected during restricted detergent use.

Time of day collected	Percent of total composite[a]	Analysis on Homogenized, Unfiltered Samples	
		$PO_4^{\equiv}$ - P mg/l	Total P mg/l
7:30 AM	3.6	3.9	4.4
8:00 AM	4.4	4.4	8.4
10:00 AM	1.3	2.8	3.2
10:30 AM	1.8	7.1	---[b]
11:00 AM	1.1	1.7	3.4
11:30 AM	2.1	2.4	5.8
1:00 PM	2.1	8.6	7.8
3:00 PM	1.2	2.2	2.8
4:30 PM	1.5	6.4	6.8
5:00 PM	6.8	6.4	14.2
5:30 PM	2.1	2.4	5.4
6:30 PM	3.3	3.8	4.6
9:00 PM	3.1	6.6	7.6
Total	34.4	---	---

[a]Volume of samples added to composite multiplied by 100 and divided by total volume of composited sample.

[b]No analysis.

Table 6. Concentrations of total phosphorus in different detergent brands.

Liquid Detergents		Concentrations in Soap Solution
Brand	Number Given to Test Neighborhood	
Dove	(23)	11 mg/l
Ivory	(8)	46 mg/l
Joy	(12)	74 mg/l
Lux	(16)	6 mg/l

Table 7. Variation in chemical constituents of raw and treated sewage samples.

Chemical Constituent	Baseline Sample			Test Sample		
	Raw	Secondary	Tertiary	Raw	Secondary	Tertiary
Carbon						
Total, mg C/l	172	96	54	288	212	202
Organic,[a] mg C/l	106	26	14	204	136	148
Inorganic, mg C/l	66	70	40	84	76	54
Inorganic (from alkalinity), mg C/l	89	92	43	102	81	74
Nitrogen						
Ammonia, μg N/l	30,000	26,000	29,000	34,000	31,000	29,000
Nitrate and nitrite, μg N/l	14	83	8	12	103	< 1
Organic N, μg N/l	12,000	5,600	200	16,000	1,600	1,000
Phosphorus						
Total, μg P/l	18,700	17,000	650	8,100	6,500	250
Filtrable orthophosphate,[b] μg P/l	9,400	12,800	41	4,700	4,200	30
Iron						
Total, μg Fe/l	480	234	125	276	128	105
Filtrable,[b] μg Fe/l	220	90	46	400	96	81
BOD_5, mg/l	360	< 20	20	590	22	< 20
pH	8.2	8.2	6.5	7.0	8.3	7.4
Alkalinity, mg/l as $CaCO_3$	370	380	94	340	335	280
Conductivity, μmhos/cm at 25°C	958	840	1,240	846	702	1,240
Suspended Solids, mg/l	195	35	11	226	25	13
Volatile Suspended Solids, mg/l	183	30	11	216	22	8.2

[a] Organic is difference between total and inorganic carbon.

[b] Filtrable is material which passes a 0.45 μ MP filter.

and pH as expected from the effect of those chemical reactions and increased the conductivity as a result of increased ions.

The most interesting comparisons were the effect of restricting dishwasher and clotheswasher use on organic carbon, BOD, and solids. Apparently because of the lack of dilution from large volume dishwasher and clotheswasher water use, the organic content (BOD and organic carbon) of the raw sewage was increased by a factor of 2 for the test sample as compared to the baseline sample.

Calculated phosphorus concentrations for different dilutions of the secondary and tertiary effluents of the two sewage samples indicate the phosphorus content was still relatively high (Table 8); this resulted from the relatively high phosphorus content in the reservoir water (Table 9). This asymptotic relationship can be clearly observed in Figure 4 where the limit for phosphorus concentration becomes equal to the reservoir concentration at infinite dilution. Therefore, if a reservoir or any water body naturally has a high phosphorus concentration, the addition of more phosphorus probably will have no effect on algal growth.[9] In Utah it would be unlikely that phosphorus would ever be limiting in aquatic systems because even the groundwater typically is about 25 μg P/l.[10]

Other analyzed nutrients seemed to be in plentiful supply in reservoir water (Table 9). The quality of the water is typical of the hard waters which drain limestone. Because the reservoir samples were collected as the spring thaw was beginning, the reservoir water was quite turbid and required extensive filtering 1) through a GF/C Whatman filter overlaid with a Millipore filter pad, and 2) through a 0.45 μ, Millipore, Type HA membrane filter.

Table 8. Calculated phosphorus concentrations in filtered reservoir water containing different sewage effluents.

Chemical Form of Phosphorus	Concentration in Reservoir Water, %	Two Percent Effluent in Reservoir Water[a] µg P/l			
		Secondary Samples		Tertiary Samples	
		Baseline	Test	Baseline	Test
Total Phosphorus	2.0	360	150	38	30
	0.8	160	80	30	27
	0.32	79	48	27	26
Filtrable Orthophosphate	2.0	280	110	25	25
	0.8	127	58	25	25
	0.32	66	38	25	25

[a]Method of calculation:

Final P Conc. = 0.02 (P Concentration in Effluent) + 0.98 (P concentration in Reservoir Water

Note: 25 µg P/l in Reservoir Water

Table 9. Chemical constituents in the First Dam reservoir water samples used for dilution water in bioassays.

Chemical Constituent	Baseline µg/l	Test µg/l
Carbon, mg C/l		
Total	45	-
Organic[a]	6	-
Inorganic	39	-
Inorganic (From Alkalinity)	40	40
Nitrogen		
Ammonia-N	50	34
Nitrate + Nitrate - N	340	230
Organic N	500	-
Phosphorus		
Total P	60	61
Filtrable[b] Orthophosphate P	60	25
Iron		
Total Fe	76	48
Filtrable[b] Fe	34	20
Alkalinity	170	170
pH	8.2	8.2
Conductance	375	354

[a]Organic is difference between total and inorganic carbon.

[b]Filtrable is material which passes a 0.45 µ MP filter.

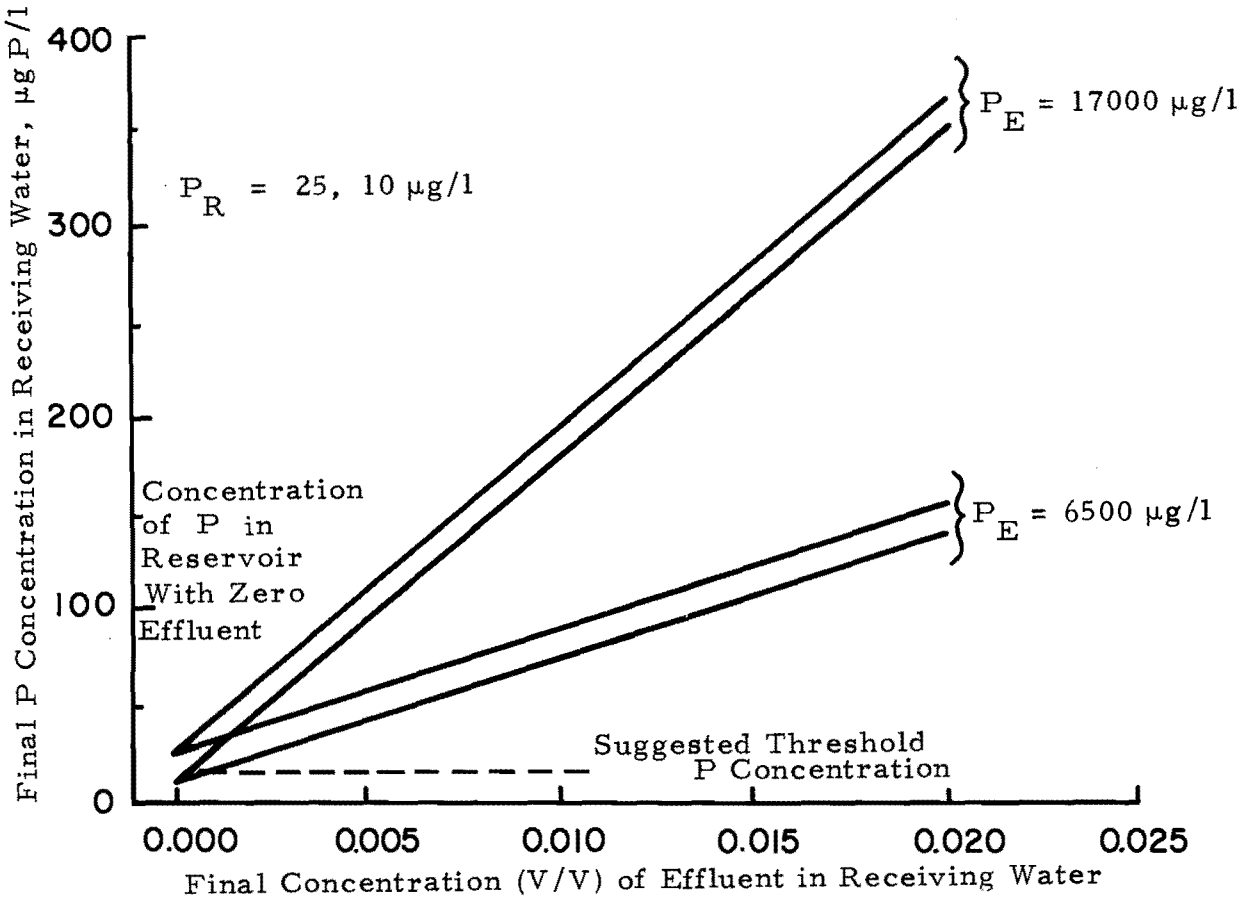

Figure 4. Effect of Dilution on the Phosphorus Concentration of a Receiving Water.

Bioassay Results

Response criteria

In the bioassays algae are added at a standard initial concentration (X_o) and allowed to grow under standard conditions.[7] Then, measurements of algal biomass are made at day 7 (X_7) and at the end of the growth cycle ($\hat{X}$). The growth cycle essentially terminates before 14 days have elapsed but generally in most studies a 21 day measurement is taken for $\hat{X}$. In this study $\hat{X}$ was taken at 18 days for the baseline sample because of scheduling problems and at 21 days for the test sample. Essentially no difference is expected due to the time difference and the measurements are treated as equivalent.

Two parameters of growth response are discussed herein, 1) maximum growth ($\hat{X}$) and mean growth rate ($\bar{\mu}_{b_7}$). Maximum growth is based upon the first and last measurements of biomass obtained during the growth cycle (18 or 21 days) and growth rate is determined as follows:

$$\bar{\mu}_{b_7} = \frac{1}{t} \ln (X_7/X_o), \text{ days}^{-1}$$

where t is time in days. The growth rate is related to the concentration of the limiting factor according to Monod[11] and as shown by Porcella et al.[12] The measured mean growth rate ($\bar{\mu}_{b_7}$) represents growth over a seven day period and was determined by making one measurement at the end of 7 days, and thus is only an estimate of the actual batch specific growth rate.

A more intuitively useful estimate of growth rate can be determined as the mean doubling time (t_d) which is the average time in hours in which the algal population doubles:

$$t_d = \frac{(\ln 2)}{\overline{\mu}_{b_7}} \quad (24 \text{ hrs/day}), \text{ hours}^{-1}$$

Doubling time decreases as the growth rate increases. Discussion in relation to sample type, dilution, and treatment will be limited to maximum and mean growth rate and the mean doubling time.

Biomass measurements

Correlation between particulate carbon (PC), optical density (OD), and suspended solids (SS) were performed to establish the relations between the individual parameters (Table 10). Originally the SS data did not correlate well with either OD or PC (data not shown here, $r = 0.80$, 0.66, respectively) but a reasonably high correlation existed between OD and PC ($r = 0.94$). Closer analysis of the data indicated that the SS measurements were unreasonably high in the samples diluted with reservoir water. Even those bioassay samples which appeared to be clear contained high concentrations of SS. This was apparently not caused by precipitation of alkaline earth carbonates on the filter paper because washing those filters with dilute acid (HCl, pH = 5) did not reduce the SS measurement. Following the acid rinse, the filters from the reservoir water dilutions were ashed at 560°C and volatile suspended solids (VSS) concentrations were determined. These VSS values compared

Table 10. Relationships between different biomass parameters for all bioassays with both Selenastrum capricornutum and Anabaena flos-aquae as test algal species.

Parameters	Number of data points	Correlation coefficient	Slope	Intercept
PC = f(OD)	136	0.94	0.0058	0.0053
PC = f(SS)	74	0.88	2.2	2.8
SS = f(OD)	74	0.88	350	3.5

reasonably well with the other parameters and thus were included in the correlations in Table 10. It was noted that the ash residue was a fine particulate inorganic material. It was surmised that this was a colloidal, mineral material from the reservoir sample which was not removed by membrane (0.45 μ MF) filtration and then aggregated or became associated with the algal cells and thus contributed to the SS measurements.

The results in Table 10 indicated that it was feasible to use PC as an estimate of biomass and Selenastrum capricornutum as a representative of other algal species in terms of relative biomass. However, correlation of response of S. capricornutum with responses of Anabaena flos-aquae indicated that considerable variability in response occurred. The correlation of final day PC measurements for both species was only 0.51 with a least squares equation as follows:

$$\text{mg PC/l } \underline{\text{A. flos-aquae}} = 0.41 \text{ mg PC/l of } \underline{\text{S. capricornutum}} + 6.1$$

Correlation between growth rate estimates indicated an inverse relationship existed ($r = -0.48$):

$$\hat{\mu}_{b_7} \text{ of } \underline{\text{A. flos-aquae}} = -0.76 \left(\hat{\mu}_{b_7} \text{ of } \underline{\text{S. capricornutum}}\right) + 0.86$$

If adequate correlation and a reasonable relationship existed between the response of the two algal species, it would be sufficient to utilize S. capricornutum as the representative of both algal species but such was not the case. Thus, both species of algae will be discussed separately,

and only $\hat{X}$ and t_d estimates based on PC measurements will be considered except where confirmation by other biomass measurements of a given data point is required.

Effects of dilution on maximum growth response ($\hat{X}$)

Dilution studies were made to detect toxicity, i.e., unusual growth as a function of dilution indicated toxicity. In these results the fraction of growth due to the dilution water alone is shown so that the actual contribution by the effluent to $\hat{X}$ can be seen independently of nutrient concentration changes in the dilution water. In the bar graphs the contribution from the dilution water is shown on top to facilitate comparison of the effluent nutrients. This is a necessary correction due to the variability in growth response obtained with the dilution waters (see Appendix C-2).

Selenastrum capricornutum. Responses shown in Figure 5 show that toxicity probably was not present in either the reservoir, baseline, or test secondary samples. The difference between baseline and test secondary samples was not significant. There was slightly more growth in the samples diluted in reservoir water than would be expected based on the growth in the deionized water, and this might have resulted from a synergistic effect of having two different nutrients limiting in the two different samples.

There was an obvious effect of tertiary treatment on the sewage samples. Growth was reduced to essentially unmeasurable levels with

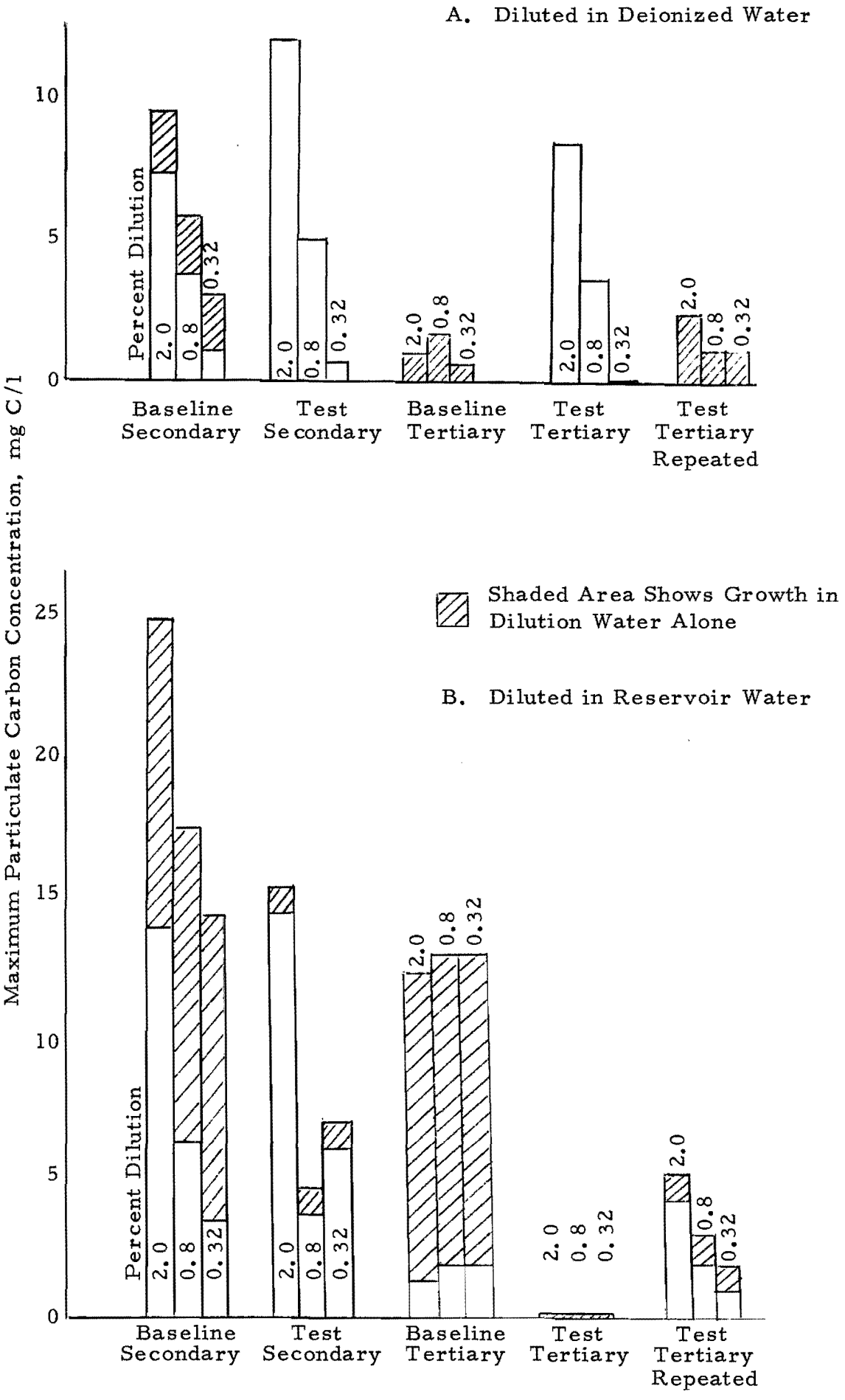

Figure 5. Dilution bioassays with Selenastrum capricornutum.

the exception being the test tertiary sample diluted in deionized water. Those deionized water results were apparently due to unknown analytical error as other biomass parameters indicated no response. Also the repeat bioassay with deionized H_2O indicated no response. Toxicity present in tertiary samples apparently caused these responses and will be discussed further in the section on spiking.

Anabaena flos-aquae. Essentially similar results were obtained with the blue-green algae although more variability in results was obtained (Figure 6). This may have been caused by the difficulty in accurately sampling filamentous species of algae. However no essential differences in baseline and test secondary samples were noted with the possible exception of the dilution of sample in reservoir water. However, considerable scatter in results was obtained in these sets of results. Also the relationship between dilution concentrations and response was confusing and not as clear as for S. capricornutum. Sampling and analytical difficulties may have been responsible for some of these results because considerable sticking of the cells to the flask walls was observed. Also it was difficult to obtain accurate samples of this filamentous organism. The results do generally support the conclusion that tertiary treatment reduces response and that there is no consistent demonstrable difference between responses to baseline and test sample.

Comparison of all two percent samples

Statistical comparison of all the two percent samples essentially confirmed the picture described in the preceding section (Table 11). The

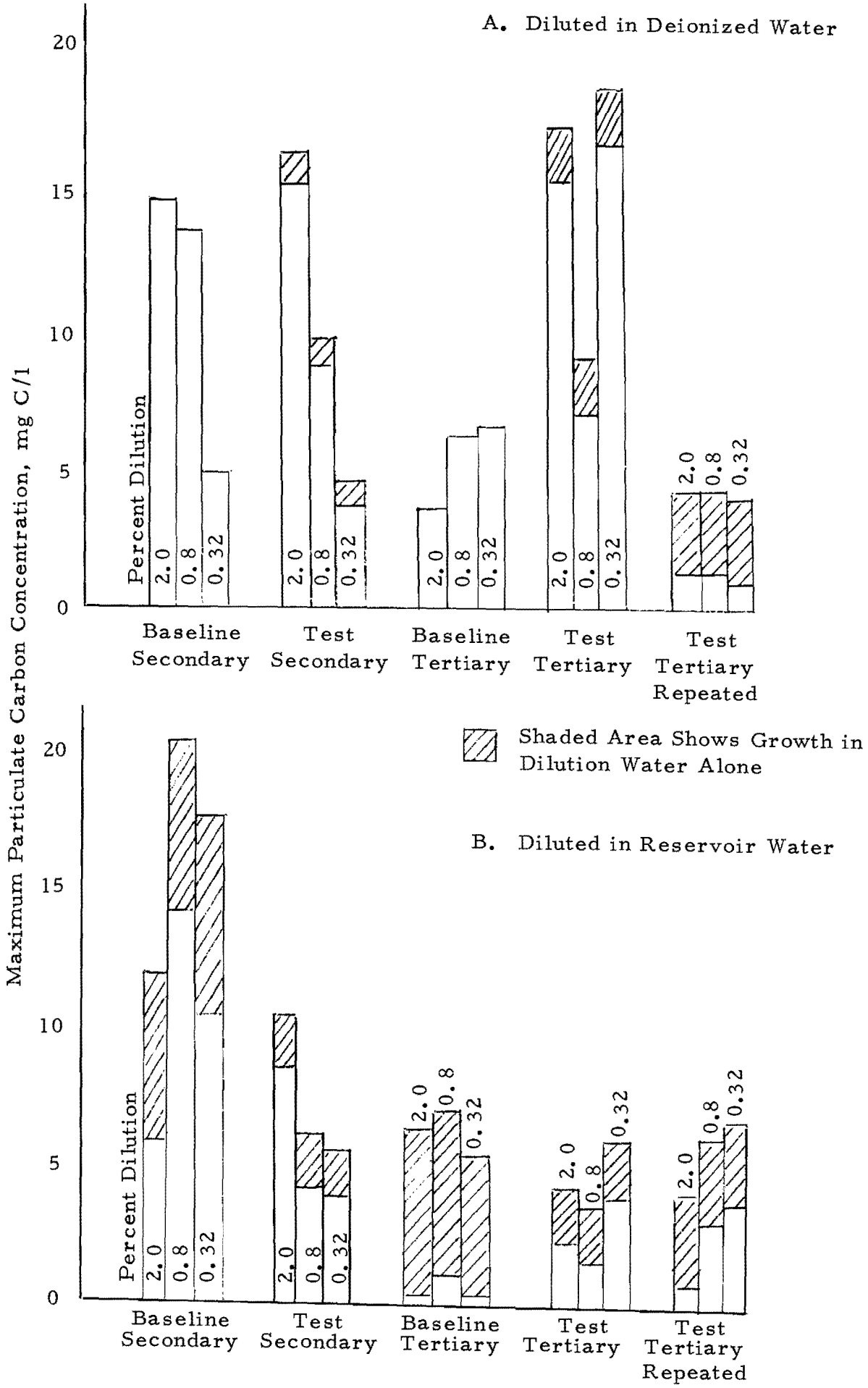

Figure 6. Dilution bioassays with Anabaena flos-aquae.

Table 11. Ranked bioassay responses of two percent samples[a] in dilution water.

Species	Dilution Water	Mean Maximum Particulate Carbon, mg/l[b]			
S. capricornutum	Deionized	TB	TT	SB	ST
		1.00	2.33	9.33	12.00
S. capricornutum	Reservoir	TT	TB	ST	SB
		5.00	12.33	15.33	24.67
Corrected[c]		4.00	1.33	14.33	13.67
A. flos-aquae	Deionized	TB	TT	SB	ST
		3.67	4.33	14.67	16.33
A. flos-aquae	Reservoir	TT	TB	ST	SB
		4.00	6.33	10.67	11.33
Corrected		3.00	0.33	8.67	5.33
S. capricornutum	Reservoir + N	TT	TB	ST	SB
		1.67	8.67	16.33	20.33
Corrected		0.67	0	15.33	9.33
S. capricornutum	Reservoir + P	TT	TB	ST	SB
		5.67	11.33	13.00	18.00
Corrected		4.67	0.33	12.00	7.00
S. capricornutum	Reservoir	TT	TB	SB	ST
	+ Fe & TE	3.33	12.00	23.33	23.67
Corrected		2.33	1.00	12.33	22.67
S. capricornutum	Reservoir + NAAM	TT	TB	SB	ST
		6.00	26.33	29.33	37.00
Corrected		5.00	15.33	18.33	36.00

[a]SB is secondary baseline, ST is secondary test, TB is tertiary baseline, TT is tertiary test (repeat assay).

[b]Underlines indicate that the values are not significantly different ($p > 0.95$).

[c]Corrected refers to the subtraction of the amount of growth due to nutrients contained in the dilution water. Only the reservoir water data were corrected as the deionized water gave relatively constant results (Appendix C-2).

comparisons were made using Duncan's multiple range test for a Split-Plot Design.[13] There were no statistical differences ($p > 0.95$) between the baseline or test samples for either secondary or tertiary samples for both bioassay species. (Note: this statement refers to data corrected for dilution water contribution to bioassay response.) However the spiking results are somewhat confusing in that the test secondary sample usually gave statistically greater bioassay response than the baseline secondary. Further experimentation would be required to explain this observation.

A comparison of all the bioassay methods for each type of sample provides more information (Table 12). The same statistical method was utilized.[13] The highest response for all samples was for the spiking addition of complete NAAM solution as would be expected. However the minimal amount of growth observed in the test tertiary bioassay was confirmed evidence of the presence of toxicity as determined from the dilution results. Although these results were obtained from the repeat bioassay, the initial bioassay confirmed that result. Almost all of the results of the test tertiary bioassay were statistically equivalent. The second highest response was obtained with either the sample alone or with iron and trace elements added, indicating that some additional response was obtained with this addition. The least response was usually obtained with S. capricornutum in deionized water while the response of A. flos-aquae in both dilution waters was also of lesser response. However, the statistical differences were not always so clear.

Table 12. Ranked effect of different assay methods on two percent samples in dilution water.[a]

Secondary		Tertiary	
Baseline	Test	Baseline	Test (repeat assay)
29.33(F)	37.00(F)	26.33(F)	6.00(F)
24.67(B)	23.67(E)	12.33(B)	5.67(D)
23.33(E)	16.33(C)	12.00(E)	5.00(B)
20.33(C)	16.33(G)	11.33(D)	4.33(C)
18.00(D)	15.33(B)	8.67(C)	4.00(H)
14.67(G)	13.00(D)	6.33(H)	3.33(E)
11.33(H)	12.00(A)	3.67(G)	2.33(A)
9.33(A)	10.67(H)	1.00(A)	1.67(C)

[a]Letters indicate assays as follows:

Selenastrum	Dilution Water
A	deionized
B	reservoir
C	reservoir + N
D	reservoir + P
E	reservoir + Fe and Trace Elements
F	reservoir + NAAM
Anabaena	
G	deionized
H	reservoir

[b]Bar indicates that the values are not significantly different ($p > 0.95$).

In summary the results in Tables 11 and 12 indicate the following conclusions: 1) no differences exist between baseline and test samples, 2) response was greater in secondary than tertiary effluents, 3) iron and trace elements appeared to stimulate slightly greater growth, and 4) the tertiary test sample was toxic.

Add back studies

Samples of various wash products were added to a solution of two percent test secondary effluent in reservoir water and the results showed no significant difference ($p > 95$ percent) in maximum growth (Figure 7). It can be concluded that the addition of the wash products to heated sewage did not affect the bioassay response. However, this result may not relate to the effects of phosphorus because hydrolysis of polyphosphates contained in Bold® may not have occurred. This might have occurred if the detergents had gone through sewage treatment. This criticism seems unlikely because as shown previously in Tables 11 and 12 the addition of orthophosphate in the spiking test had no effect on response. In essence this experiment only demonstrated what was already known, that phosphorus would not stimulate growth in secondary effluent.

Rates of Growth

Doubling times were measurable only for the 2 percent samples because the methods for measuring biomass were not sensitive enough to obtain good measurements in the low nutrient bioassays after 7 days of growth. The results indicate that S. capricornutum grew faster than

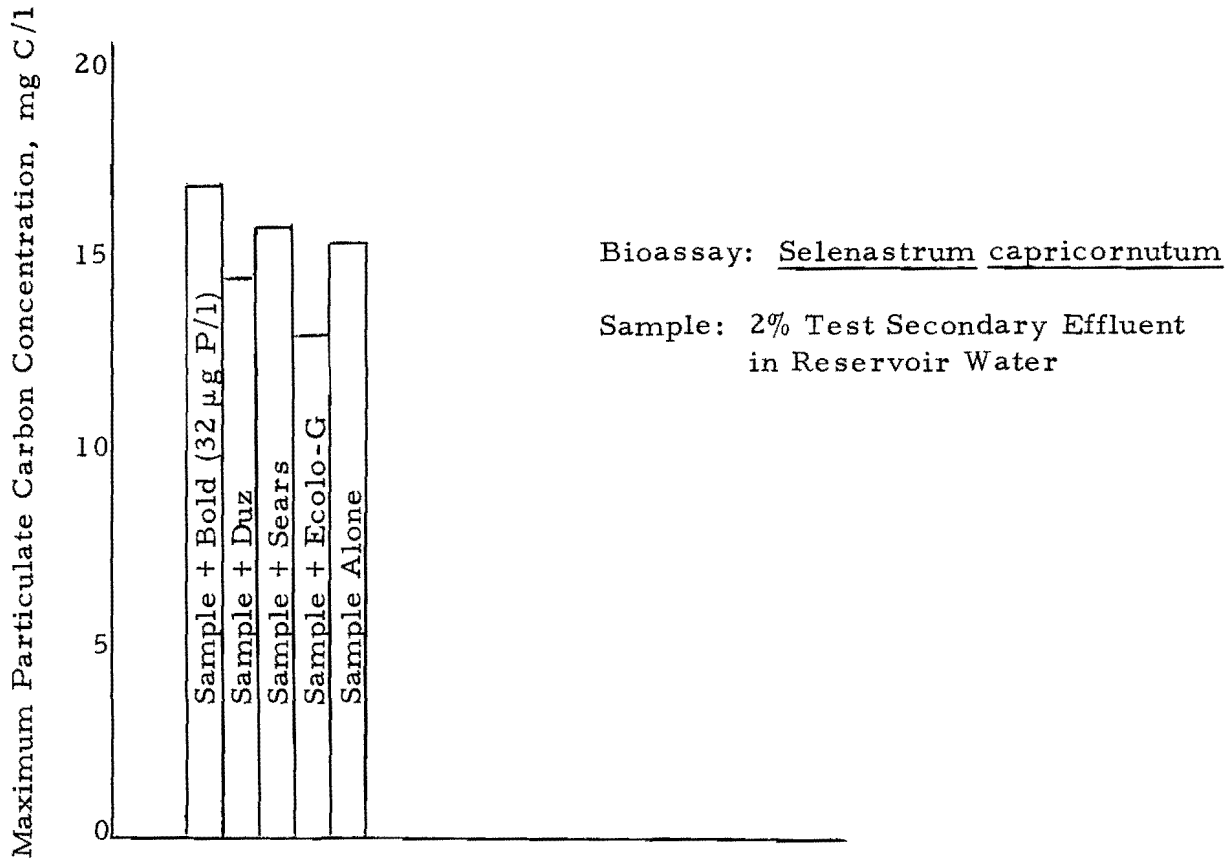

Figure 7. Add back bioassays measuring wash product effect on maximum growth.

A. flos-aquae (Table 13). Other conclusions such as whether secondary or tertiary effluents had greater effect on response, or whether the baseline or test samples were more stimulatory, were not clear because of the scatter in the data.

The most important aspect of these studies is in defining the limiting nutrient. This is not a simple matter especially when the results are later applied from laboratory studies to natural conditions. Previously almost all such definitions were based primarily on maximum growth measurements as presented above. Goldman[14] has used ^{14}C uptake rate measurements in field bioassays and Pearson[15] has advocated growth rate measurements for laboratory bioassays as being more closely related to natural conditions than maximum growth. This seems logical because the organism that replaces itself the fastest in the face of predation, etc., will become dominant.

The effects of various spikes on growth rate expressed as the time in hours necessary to double the amount of algae are summarized in Table 14. The lowest doubling times (fastest growth rates) were invariably associated with the addition of chelated iron and trace elements either alone or in the complete medium (NAAM). Nitrogen or phosphorus alone did not change the growth rate in comparison with the control (no addition). This indicated that iron or one of the trace elements was actually limiting and not phosphorus or nitrogen. The minimum observed doubling time (15.9 hours) corresponds to a mean specific growth rate of 1.05 $days^{-1}$ a value not too much lower than reported values of 1.20 $days^{-1}$ for the

Table 13. Average doubling times for two percent effluent diluted in deionized or reservoir water.

	t_d, hours			
	Secondary		Tertiary	
	Baseline	Test	Baseline	Test
Selenastrum capricornutum				
Deionized Water	19.6	18.5	--[b]	20.5[a]
Reservoir Water	18.5	18.0	17.8	19.6
Anabaena flos-aquae				
Deionized Water	63.3	63.3	--[b]	260.5, 84.5[a]
Reservoir Water	260.5	84.5	75.6	69.3[a]

[a]Repeat Bioassay.

[b]Not possible to estimate doubling times.

Table 14. Average doubling times of Selenastrum capricornutum in spiked samples.

Type of Addition	Avg. Doubling Time in Hours Over the Initial Growth Period (0-7 Days) Two Percent Secondary Effluent in Reservoir Water	
	Baseline	Test
Nitrogen	19.9	-
Phosphorus	22.0	-
Iron & Trace	15.9	17.0*
NAAM	16.0	16.5
No Addition	18.5	18.0

*Results enclosed in box are significantly different using a t-test from all other doubling time estimates at $p > 95$ percent. Dash indicates that the growth rate was too low to be measured.

maximum specific growth rate batch under the same environmental conditions for the test alga, Selenastrum capricornutum.[12]

The observed maximum growth values did not present as clear a picture as growth rate. However, those results generally supported the statement that nitrogen and phosphorus were not limiting in the secondary effluents and that some other factor was.

Conclusions

1. Restriction of detergent use by preventing use of dishwashers and clotheswashers led to a 57 percent reduction in total phosphorus content of raw sewage.
2. BOD_5 and organic carbon content in the test sample was double the concentration in the baseline sample, presumably due to lack of dilution from dishwasher and clotheswasher use.
3. The minimum concentration of phosphorus in any body of water receiving sewage effluent will be the natural phosphorus concentration.
4. Bioassay response was greater in secondary effluents than tertiary for both baseline (collected before detergent restriction) and test (collected after detergent restriction) samples.
5. No significant difference in bioassay response was detectable between the baseline and test secondary effluents.
6. No significant difference in bioassay response was detected between the baseline and test tertiary effluents. The response was extremely

low thus precluding firm conclusions about tertiary effluents, and toxicity was observed in the test tertiary sample.

7. Spiking studies indicated that phosphorus was not limiting in any of the samples. Most probably iron or one of the trace elements limited growth.

8. Addition of wash products to test secondary effluent did not significantly affect the bioassay results.

9. For the dilution waters utilized in these bioassays, the results indicate that restriction of phosphorus use in detergents would have no effect on the algal growth response to secondary effluents.

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Acknowledgments

Considerable credit should be given to the efforts and experience of Mr. Peter Cowan and to Mr. Harold Jones, Mr. Steven Nielson, and Mr. Richard Svetich for their willingness to work unusual hours in addition to their regular work. Besides the laboratory work and sampling the sewage, these four distributed questionnaires, the free gifts, and explained the purpose of the project to homeowners during the day and on weekends and evenings.

Mr. Ray C. Hugie, Logan City Engineer, and his staff made suggestions and generously lent a weir and opened the blueprint file for our use.

Also Mrs. E. J. Middlebrooks, Mrs. Paul Koenig, and Mrs. J. W. Jackson helped very much in gaining the cooperation of the neighborhood. They phoned all the neighbors, asking them to meet at Mrs. Middlebrooks' house for refreshments so that slides could be shown and further questions answered.

The support and suggestions of Dr. Jay M. Bagley, Director of the Utah Water Research Laboratory, were appreciated. The enthusiastic support and willingness to handle rush typing jobs by the secretaries, Betty Hansen, Barbara South, and Vicki Smith makes the arduous task of preparing a report a pleasant experience.

Appendix A-1

April 16, 1971

Dear Householders:

Most of you are aware of the many environmental problems which have arisen in our country and in all modern, industrial societies. These problems occur because the natural environment is no longer able to assimilate the large amounts and exotic kinds of materials which we discharge to them. It is now technology's turn to begin developing solutions for these problems, many of which have resulted from technological advances. However, to obtain solutions, accurate information about the sources, kinds, and amounts of specific pollutants are needed. As part of this search to quantify pollutants discharged to the environment, the Utah Water Research Laboratory, College of Engineering, Utah State University is beginning a study to define the relative proportions of phosphates from detergent and non-detergent sources in residential areas. This study will require your cooperation.

Before explaining how this will be done, a little background information on how phosphates affect our environment might be helpful. Phosphorus in the form of phosphates is one of many nutrients (such as nitrogen, iron, and potassium) which is used by all plants and animals. Phosphorus forms about 1% by weight of the average human; and, obviously it is not a toxic substance. But it can lead to problems. This is because most natural bodies of water are low in phosphorus, and the phosphorus and other nutrients which man adds through his many activities (agricultural fertilizers, sewage, mining, and other wastes) can do in lakes what fertilizer can do in your lawn: make them greener. In lakes this is usually seen as an increased growth or a bloom of algae (small green plants). This bloom can become so extensive that scums appear on the surface of the lake, the oxygen is used up, and fish die. The process of increasing nutrients in lakes and streams is called "eutrophication" and when the actions of human societies lead to overfertilization of lakes it is called "cultural eutrophication."

Therefore this study will be focused on the contribution of detergents to the phosphorus concentration in sewage. This is because in lakes around municipalities most of the phosphorus going into those lakes is from sewage and about half of the sewage phosphorus comes from detergents. To measure accurately this detergent phosphorus contribution, we need to study an area of Logan which can be experimentally isolated from any other sources of sewage.

This northeastern corner of Logan where you reside meets this requirement and has other advantages: it is a new area of town and the sewer pipes are well constructed, minimizing infiltration from the groundwater. Also, people such as you have the most modern appliances and tend to be high users of detergents, soaps, and cleaners. Moreover you have the most at stake and the most interest in preserving our environment, and we assumed that we could get complete cooperation. The first reasons become valueless without the cooperation of the neighborhood.

This cooperation will cause some slight inconvenience to residents, but we will minimize this as much as possible. The inconvenience involves not using your automatic dishwashers and clothes washers at all during a period of three days (Mon.-Wed., April 26-28, 1971; see attached instructions). In turn we will provide you with a liquid detergent for use in the kitchen and a three-day supply of paper plates and cups. If there is any particular inconvenience caused by not using your clothes washer, we will repay you for laundry services or set up an account with a diaper service to handle special laundry needs. We are open to other suggestions about ways to minimize any problems. In this way we hope that the inconvenience will be minimal and also that you will feel that you have contributed materially to this study.

This study is one of those few which provides results which have immediate impact on the public, and we are pleased that we can involve the community. Because the public usually is affected greatly by such results, it is important that they be involved and that the results truly reflect their habits. This is why we need your cooperation. Although your cooperation requires that you refrain from using the dishwasher, clothes washer, and from heavy cleaning (washing floors, walls, etc.), in every other respect you are to observe your normal routine.

We are planning to have an introductory meeting in Room EC 101 in the Engineering Building at Utah State University (across the street from the Fine Arts Center) at 7:30-8:00 P. M. on Thursday, April 22, where we will show slides, accept suggestions, and answer questions. We would also like you to fill out the questionnaire which is enclosed (no personal information involved). Those of you who cannot make the meeting will be contacted personally. On Thursday or Friday, April 29 or 30, we will collect the questionnaire and ask you to sign an affidavit saying you did not use your dishwasher or clothes washer during April 26 through 28, 1971. We sincerely hope that you all will be interested in this study and that we have 100 percent cooperation. We are looking forward to seeing as many of you as possible at our meeting on April 22.

Sincerely yours,

Donald B. Porcella

Donald B. Porcella
Assistant Professor

DBP:bjh

Attachments:

1) Instruction Sheet
2) Questionnaire
3) Affidavit
4) Laundry Claim Sheet

Questionnaire

(If you do not wish to answer or do not know the answer, place a dash in the space.)

1. Inhabitants of house

   Number of adults _____
   Number of children (under 18) _____

2. Number of water use facilities

   Toilets _____
   Bathroom sinks _____
   Other sinks _____
   Bath or shower _____
   Dishwasher _____
   Clotheswasher _____
   Garbage grinder _____

3. Does your house use a water softener? Yes ____ No ____

4. Does the water softener treat all the water or just the hot water (circle answer)?

5. How much detergent do you normally use? _____ lb/week

6. Would you be interested in participating further in this study if we provided a detergent substitute? Yes ____ No ____

Affidavit Logan, Utah 84321

I, _____ (full name), state that neither a dishwasher nor a clotheswasher were used on the premises during the period April 26 through April 28, 1971.

Signed _____
Street Address _____
Date _____

Instruction Sheet

1. Do not use any dishwasher or clotheswasher on Monday, Tuesday, or Wednesday (April 26, 27, 28). Also refrain from heavy cleaning (scrubbing floors, washing walls, etc.). Sampling will be completed at 1 a.m., Thursday, April 29, and after that time everything can return to normal.

2. In all other respects do what you would normally do during that week. Do not worry about the use of any other appliances (garbage grinders, etc.) or use of hand soaps. These each supply a minor part of the total phosphorus in sewage and so just use them as usual.

3. It would be best if you taped your dishwasher and clotheswasher doors closed so that there will be no danger of absentmindedly using them. Also place your dishwasher and clotheswasher detergents in a box in the garage or some other out-of-the-way place for the same reason.

4. Attached is a form for claiming laundry expenses.

If there is any problem about laundering, please fill out this form and mail in the attached, pre-addressed and stamped envelope. A check covering your laundry expenses will be mailed to you.

Name _____

Address _____

Commercial Laundry Expenses during April 26-28, 1971

Total Cost _____ (Attach Receipt)

Name of laundry used _____

PLEASE MAIL THIS FORM IN THE ATTACHED PRE-ADDRESSED AND STAMPED ENVELOPE TO: MR. PETER COWAN, UTAH WATER RESEARCH LABORATORY, UTAH STATE UNIVERSITY, LOGAN, UTAH 84321.

Appendix A-2

As my part in protecting the environment, I will cooperate with the Utah Water Research Laboratory Study and abide by the instructions provided for the period April 26-28, 1971.

Sign-up Sheet

Name	Address	Date

Appendix A-3

INSTRUCTIONS

1. DO NOT USE YOUR DISHWASHER OR CLOTHESWASHER ON MONDAY, TUESDAY, OR WEDNESDAY OF THIS WEEK. Do not use any of those detergents which you normally use in the dishwasher or clotheswasher. We suggest placing these detergents out in the garage.

2. Attached are a postage stamp and also some "peel-stickers" for sealing your dishwasher and clotheswasher. This will help remind you.

3. We will pick up the questionaire and affadavit on Thursday or Friday. Also we will provide you with a summary of results in a few weeks.

THANK YOU

Reminder

Remember to seal off your dishwasher and clotheswasher and place your boxes of detergents in a box in the garage so that there will be no danger of your using them during Monday, Tuesday, or Wednesday (April 26-28). Thank you.

Donald B. Porcella
Utah Water Research Laboratory
Phosphorus in Detergents Study

UTAH STATE UNIVERSITY · LOGAN, UTAH 84321

UTAH WATER RESEARCH LABORATORY

COLLEGE OF ENGINEERING

May 26, 1971

Mr. and Mrs. John Doe
Utah Water Research Laboratory
Utah State University
Logan, Utah 84321

Dear Mr. and Mrs. Doe:

Last month you participated in a study of the effects of hard detergents on phosphorus in sewage. Our analyses have shown that the phosphorus concentration in sewage decreased from 17 mg/1 to 5 mg/1, or a reduction to about 30% of the original level. It can be concluded that about 70% of the phosphorus came from the hard detergents.

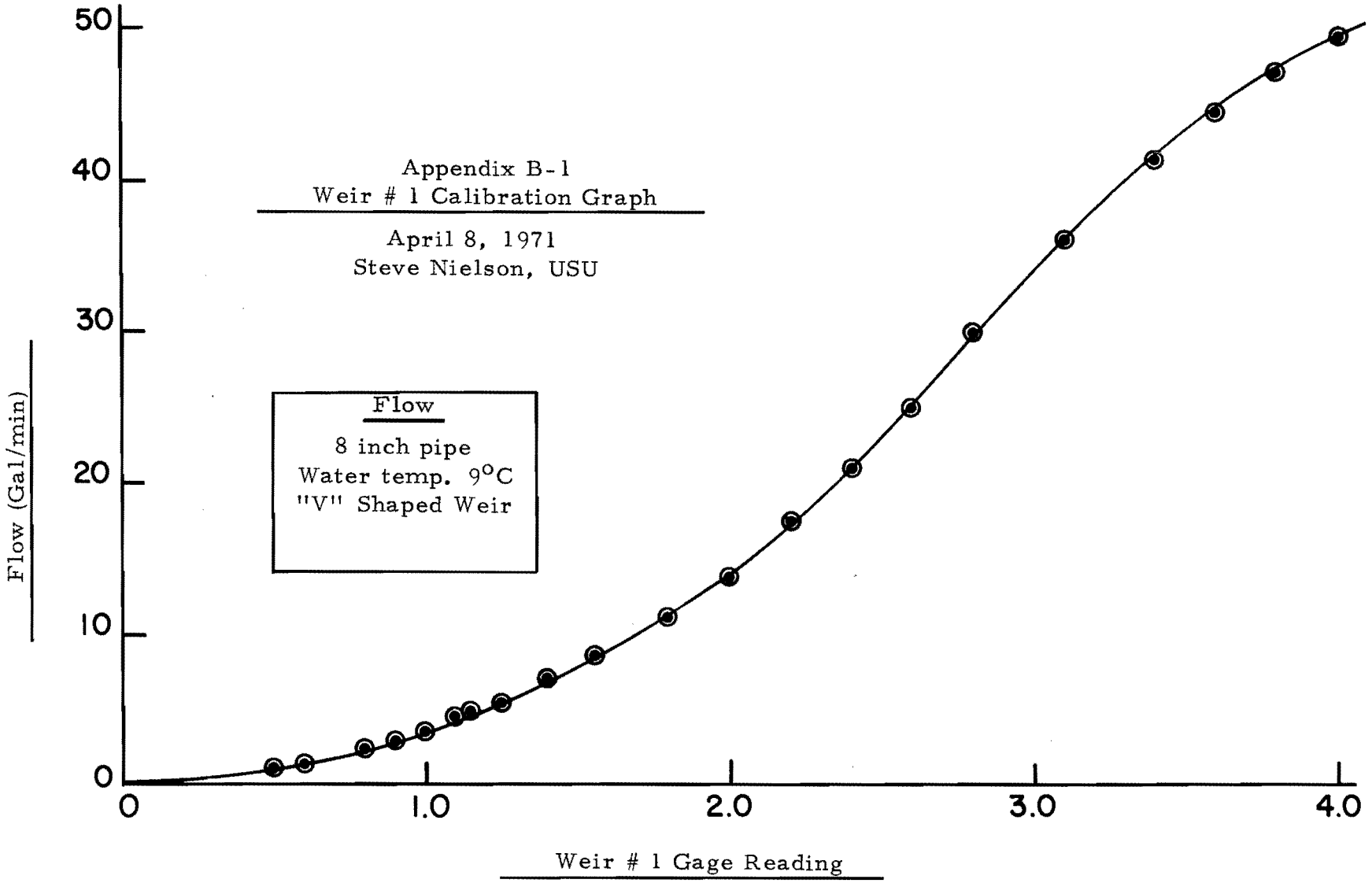
Your neighborhood is probably typical of a suburban community and this may explain the somewhat higher percentage than normally expected from cities (about 50% from detergents). Further analyses of the detergents are continuing and when these results are available, it may be possible to publish them in the newspaper. Further conclusions about the effects of phosphorus or the value of changing detergents cannot really be made until these other analyses are completed.

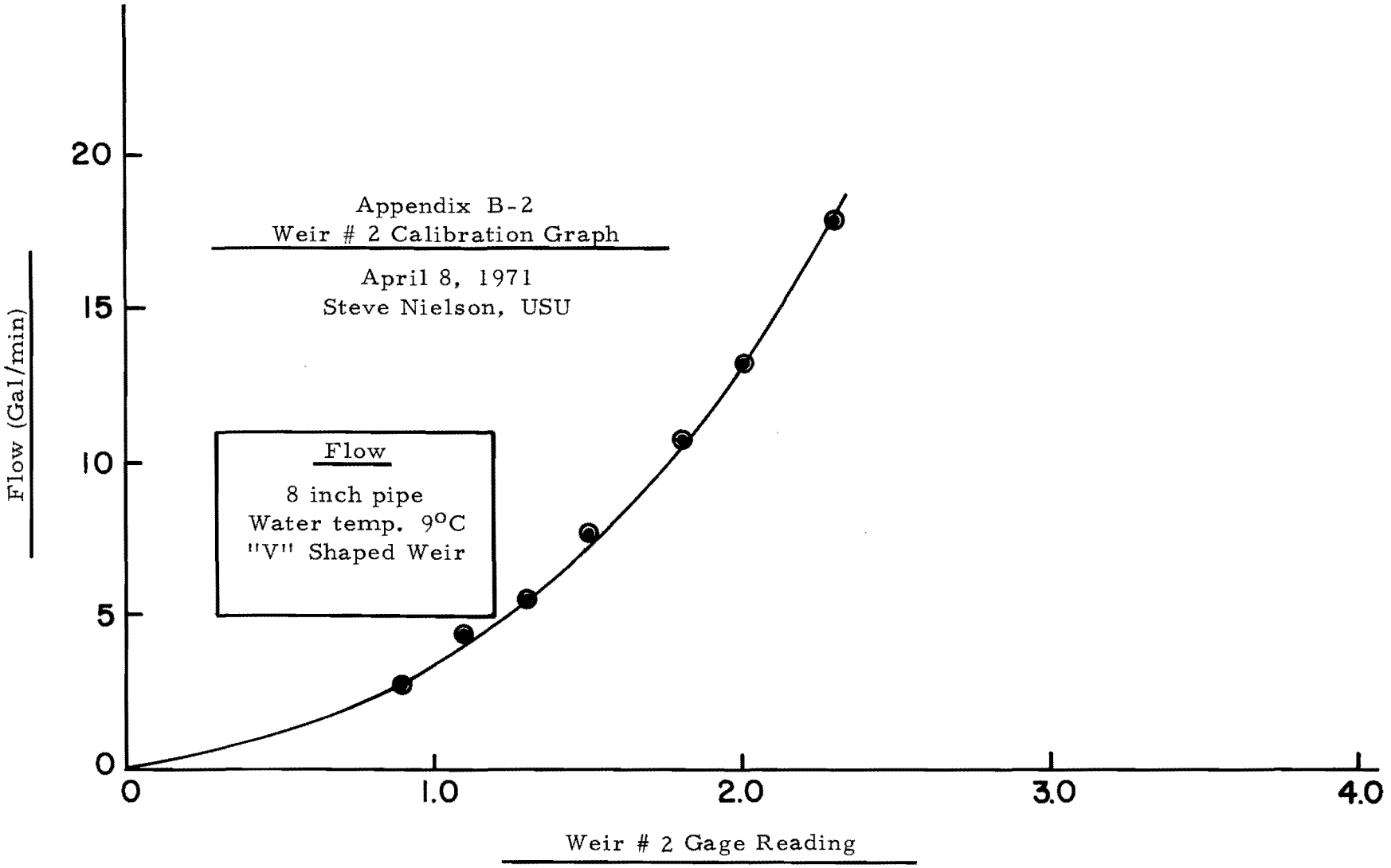
However, none of these results would be possible without your cooperation. Of the 59 households in your neighborhood, 59 returned questionnaires and affadavits, and we had essentially 100% cooperation. However, a few people noted that due to confusion they were unable to avoid using their washers entirely. We feel that these few did not have a significant effect on the overall results. We would like to thank you all for your great support and interest.

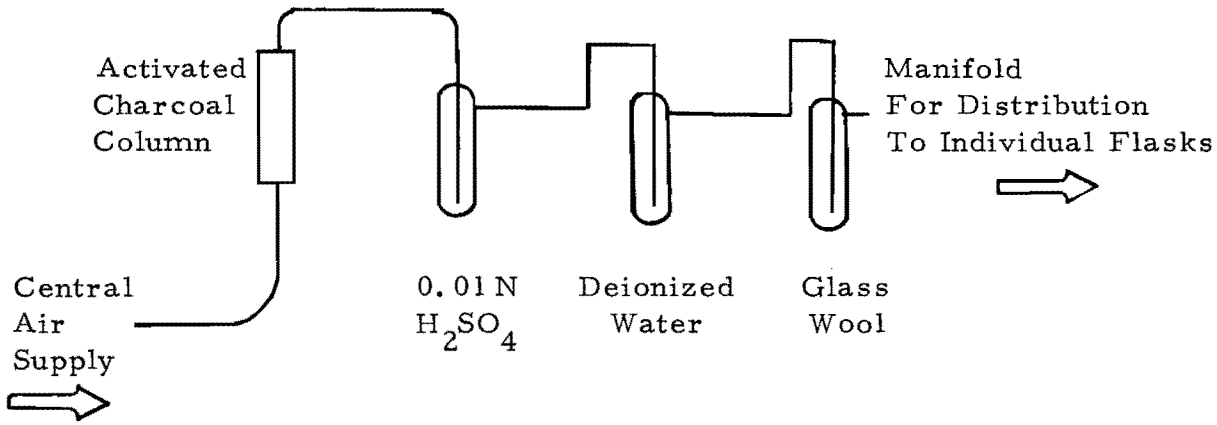
Sincerely yours,

Donald B. Porcella
Assistant Professor

DBP:vle


Appendix B-1
Weir # 1 Calibration Graph
April 8, 1971
Steve Nielson, USU
Flow
8 inch pipe
Water temp. 9°C
"V" Shaped Weir
Flow (Gal/min)
50
40
30
20
10
0
0
1.0
2.0
3.0
4.0
Weir # 1 Gage Reading


Appendix B-2
Weir # 2 Calibration Graph
April 8, 1971
Steve Nielson, USU
Flow
8 inch pipe
Water temp. 9°C
"V" Shaped Weir
Flow (Gal/min)
0
5
10
15
20
0
1.0
2.0
3.0
4.0
Weir # 2 Gage Reading



Appendix B-3. Schematic of Aeration Scheme.

Appendix C-1. Bioassay measurements for each flask.

Sample Bottle	Sample Description	Baseline Sample							Test Sample						
		7-day Analysis			18-day Analysis				7-day Analysis			21-day Analysis			
		pH	O. D.	PC	pH	O. D.	PC	SS	pH	O. D.	PC	pH	O. D.	PC	SS
A. Selenastrum capricornutum growth in secondary effluent diluted in deionized water.															
1		8.3	.042	3	7.1	.072	8		9.5	.050	1	8.1	.093	14	
2	2%	8.6	.035	3	7.2	.070	10	18.1	9.8	.050	6	8.1	.096	12	22.4
3		8.8	.045	4	7.1	.075	10		10.0	.050	7	8.1	.096	10	
4		7.8	.013	1	7.1	.032	4		9.0	.020	3	8.0	.045	5	
5	.8%	7.2	.011	-1	7.1	.027	7	9.6	9.1	.020	2	8.0	.048	5	24.2
6		8.4	.013	0	7.0	.030	6		8.9	.012	-	8.0	.040	5	
7		7.7	.005	-2	6.7	.018	3		8.2	.005	1	8.0	.017	1	
8	.32%	7.9	.007	-1	6.7	.017	3	4.9	8.0	.003	0	8.0	.016	-1	7.1
9		6.7	.007	0	6.7	.013	3		8.1	.005	0	7.9	.017	2	
B. Selenestrum capricornutum growth in secondary effluent diluted in reservoir water.															
19		9.1	.040	7	8.8	.133	26		9.0	.020	8	8.7	.118	16	
20	2%	9.2	.040	4	8.8	.133	21	50.5	9.1	.040	6	8.7	.118	16	36.8
21		9.3	.042	3	8.8	.138	27		9.0	.020	3	8.6	.100	14	
22		9.1	.016	5	8.8	.092	17		8.9	.020	7	8.7	.046	5	
23	.8%	8.6	.012	1	8.8	.094	16	36.4	8.9	.010	1	8.7	.072	4	21.9
24		9.1	.016	0	8.8	.093	19		8.9	.015	3	8.7	.030	5	
25		8.7	.020	5	8.8	.085	16		8.8	.025	6	8.8	.047	7	
26	.32%	8.9	.020	2	8.9	.080	15	29.0	8.8	.020	5	8.8	.047	7	19.3
27		8.9	.022	3	8.9	.078	12		8.8	.020	6	8.7	.044	7	
C. Selenastrum capricornutum growth in tertiary effluent diluted in deionized water.															
37		6.9	.002	-2	7.6	.007	1		9.0	.048	8	8.4	.091	8	
38	2%	7.4	.007	-2	7.2	.008	1	5.4	9.0	.045	6	8.3	.090	5	12.6
39		7.4	.004	-2	7.2	.009	1		9.2	.050	7	8.3	.091	12	
40		7.3	.003	-2	7.0	.008	1		8.5	.028	4	8.2	.040	2	
41	.8%	7.4	.005	-3	7.0	.008	2	3.3	8.4	.025	3	8.2	.035	4	5.1
42		7.0	.006	-3	7.2	.007	2		8.3	.030	3	8.2	.040	5	
43		6.7	.002	-1	6.5	.007	1		8.2	.010	2	8.2	.009	0	
44	.32%	6.7	.005	-3	6.7	.006	1	1.6	8.2	.010	1	8.2	.009	0	3.5
45		6.2	.003	-2	6.7	.005	0		8.2	.010	0	8.2	.010	0	
D. Selenastrum capricornutum growth in tertiary effluent diluted in reservoir water.															
55		8.6	.028	7	8.6	.049	9		8.7	.005	8	8.7	.001	-2	
56	2%	8.5	.022	5	8.7	.054	12	17.0	8.7	.005	1	8.7	.000	-3	0.7
57		8.4	.024	--	8.7	.055	16		8.7	.005	1	8.8	.000	-2	
58		8.7	.026	6	8.7	.065	14		8.8	.005	4	8.7	.002	-5	
59	.8%	8.5	.023	4	8.7	.070	13	14.9	8.8	.005	1	8.7	.005	-1	0.5
60		8.8	.026	4	8.7	.060	11		8.7	.005	1	8.7	.006	-5	
61		8.4	.021	6	8.7	.060	15		8.8	.005	4	8.7	.009	-2	
62	.32%	8.8	.020	3	8.8	.055	12	15.5	8.8	.005	2	8.7	.011	0	2.1
63		8.5	.020	2	8.8	.062	11		8.8	.003	3	8.8	.006	-2	

Appendix C-1. Continued.

Sample Bottle	Sample Description	Baseline Sample							Test Sample						
		7-day Analysis			18-day Analysis				7-day Analysis			21-day Analysis			
		pH	O.D.	PC	pH	O.D.	PC	SS	pH	O.D.	PC	pH	O.D.	PC	SS
E. Selenastrum capricornutum growth in tertiary effluent diluted in deionized water (repeat bioassay).															
37									7.1	.001	--	7.8	.028	4	
38	2%								7.2	.000	5	7.8	.011	1	6.8
39									7.1	.001	0	7.9	.012	2	
40									7.1	.000	2	7.6	.027	3	
41	.8%								7.0	.000	3	7.7	.020	2	6.6
42									7.1	.000	2	7.6	.007	0	
43									7.0	.000	0	7.2	.013	0	
44	.32%								7.0	.000	0	7.3	.012	0	5.7
45									7.0	.000	1	7.3	.011	3	
EE. Selenastrum capricornutum growth in tertiary effluent diluted in reservoir water (repeat bioassay).															
55									8.4	.000	0	8.6	.026	0	
56	2%								8.5	.000	0	8.5	.030	9	11.2
57									8.5	.000	0	8.6	.030	6	
58									8.4	.001	0	8.6	.030	3	
59	.8%								8.4	.001	7	8.7	.030	5	12.2
60									8.4	.002	1	8.7	.028	1	
61									8.4	.003	2	8.7	.028	1	
62	.32%								8.4	.003	5	8.7	.032	3	14.9
63									8.4	.002	7	8.6	.032	2	
F. Anabaena flos-aquae growth in secondary effluent diluted in deionized water.															
10		8.5	.025	2	8.9	.085	15		8.9	.018	2	8.8	.080	16	
11	2%	8.4	.020	1	8.9	.090	14	35.8	9.5	.035	6	8.2	.070	17	51.7
12		8.3	.025	5	8.9	.095	15		9.0	.005	0	9.0	.100	16	
13		8.5	.030	3	8.5	.085	13		9.0	.007	0	8.0	.030	7	
14	.8%	8.2	.020	3	8.4	.065	12	39.6	8.8	.000	0	8.3	.050	11	25.7
15		8.4	.030	2	8.6	.075	16		9.3	.020	2	7.9	.045	11	
16		6.9	.006	-1	8.1	.035	5		8.4	.000	-1	8.8	.030	5	
17		6.8	.003	-1	8.2	.035	5	10.2	8.5	.000	2	8.2	.003	1	9.2**
18		6.9	.001	-3	8.3	.035	5		9.0	.010	2	7.9	.035	8	
G. Anabaena flos-aquae growth in secondary effluent diluted in reservoir water.															
28		8.8	.018	3	8.4	.050	12		9.0	.010	3	8.7	.060	13	
29	2%	8.8	.028	2	8.6	.040	9	30.1	8.8	.005	4	8.7	.055	9	32.1
30		8.8	.022	-3	8.6	.040	13		9.0	.010	-2	8.7	.055	9	
31		8.7	.060	11	8.8	.095	22		8.8	.010	0	8.7	.030	8	
32	.8%	8.8	.050	8	8.6	.035	13	43.6	8.8	.010	0	8.6	.023	4	18.5
33		8.7	.043	4	8.5	.065	25		8.8	.015	1	8.6	.030	6	
34		8.5	.030	6	8.7	.090	16		8.7	.030	2	8.6	.018	8	
35	.32%	8.6	.035	3	8.8	.090	20	43.6	8.7	.015	0	8.6	.024	3	7.7
36		8.5	.028	5	8.6	.070	16		8.7	.020	0	8.6	.025	6	

Appendix C-1. Continued.

Sample Bottle	Sample Description	Baseline Sample 7-day Analysis pH	O. D.	PC	Baseline Sample 18-day Analysis pH	O. D.	PC	SS	Test Sample 7-day Analysis pH	O. D.	PC	Test Sample 21-day Analysis pH	O. D.	PC	SS
H. Anabaena flos-aquae growth in tertiary effluent diluted in deionized water.															
46		7.1	.010	-2	7.2	.018	4		9.0	.015	-1	8.9	.165	21	
47	2%	7.2	.010	-1	7.2	.015	3	5.9	9.0	.025	4	8.8	.090	11	30.2
48		7.2	.011	-1	7.2	.017	4		9.1	.012	-1	8.6	.120	20	
49		7.0	.006	-2	6.9	.022	5		9.3	.030	0	8.5	.095	--	
50	.8%	7.0	.009	-2	7.1	.022	7	7.6	8.4	.020	1	8.6	.055	8	16.3
51		7.7	.010	-1	7.0	.025	7		8.8	.040	0	8.4	.075	10	
52		7.2	.007	-3	7.1	.022	7		9.2	.035	2	8.5	.090	18	
53	.32%	7.4	.008	-2	7.3	.025	6	8.8	8.6	.040	0	8.4	.058	6	41.4
54		7.1	.008	-2	7.0	.021	--		9.2	.038	1	10.0	.190	32	
I. Anabaena flos-aquae growth in tertiary effluent diluted in reservoir water.															
64		8.5	.003	5	8.5	.029	9		8.6	.000	-3	8.6	.012	7	
65	2%	8.6	.001	1	8.5	.038	6	8.4	8.8	.020	-5	8.7	.011	3	3.1
66		8.4	.002	0	8.5	.020	4		8.8	.020	-2	8.6	.010	3	
67		8.4	.003	3	8.4	.015	7		8.6	.020	0	8.6	.012	5	
68	.8%	8.4	.003	1	8.5	.038	8	7.0	8.7	.035	3	8.6	.014	0	5.7
69		8.4	.003	0	8.5	.030	6		8.7	.001	2	8.6	.014	3	
70		8.4	.003	2	8.6	.028	4		8.6	.038	1	8.6	.012	8	
71	.32%	8.4	.002	-1	8.5	.022	5	8.3	8.6	.020	2	8.6	.011	6	4.9
72		8.4	.002	--	8.5	.050	7		8.6	.020	-2	8.6	.017	4	
J. Anabaena flos-aquae growth in tertiary effluent diluted in deionized water (repeat bioassay).															
46									7.9	.002	0	7.5	.024	4	
47	2%								8.0	.003	2	7.8	.030	6	14.6
48									7.8	.003	3	7.6	.026	3	
49									8.1	.009	2	7.2	.019	6	
50	.8%								8.0	.008	8	7.6	.035	5	11.4
51									7.9	.005	0	7.6	.012	2	
52									8.3	.008	1	7.7	.040	5	
53	.32%								8.3	.005	0	7.6	.020	2	8.3
54									7.9	.007	0	7.6	.011	5	
JJ. Anabaena flos-aquae growth in tertiary effluent diluted in reservoir water (repeat bioassay).															
64									8.6	.002	2	8.7	.030	5	
65	2%								8.6	.001	4	8.5	.020	5	8.3
66									8.6	.001	1	8.6	.034	2	
67									8.7	.000	4	8.6	.040	5	
68	.8%								8.7	.000	3	8.6	.038	7	20.3
69									8.7	.000	2	8.7	.038	6	
70									8.7	.000	6	8.7	.028	5	
71	.32%								8.7	.000	0	8.7	.036	8	13.7
72									8.7	.001	0	8.7	.033	7	

Appendix C-1. Continued.

Sample Bottle	Additions to Sample Diluted	Baseline Sample							Test Sample						
		7-day Analysis			18-day Analysis				7-day Analysis			21-day Analysis			
		pH	O.D.	PC	pH	O.D.	PC	SS	pH	O.D.	PC	pH	O.D.	PC	SS
K. Spiking studies: Selenastrum capricornutum growth in secondary effluent diluted to 2 percent in reservoir water plus added specific nutrients.															
1		8.5	.017	5	8.6	.110	20		9.0	.010	2	8.6	.118	14	
2	NH_4-N	8.6	.020	2	8.6	.115	15	48.5	8.9	.008	-2	8.5	.130	18	47.1
3		8.6	.012	2	8.6	.120	26		8.8	.007	-1	8.6	.110	17	
4		8.8	.027	4	8.6	.115	19		8.9	.008	0	8.6	.095	12	
8	PO_4-P	8.8	.022	-1	8.6	.120	19	53.5	8.9	.005	-2	8.6	.090	13	36.6
10		8.8	.022	2	8.6	.110	16		9.0	.008	-2	8.6	.090	14	
11	Trace	8.7	.092	15	8.6	.150	24		9.2	.085	8	8.6	.160	23	
12	and	8.7	.094	13	8.6	.160	23	51.8	9.3	.082	7	8.6	.160	22	54.3
13	Iron	8.7	.094	11	8.6	.150	23		9.3	.086	10	8.6	.170	26	
14		8.8	.102	14	8.6	.180	27		9.9	.102	7	8.7	.250	40	
15	NAAM	8.8	.104	12	8.6	.185	31	87.5	9.8	.106	14	8.6	.240	35	78.7
16		8.9	.108	11	8.6	.180	30		9.8	.104	9	8.6	.240	36	
L. Spiking studies: Selenastrum capricornutum growth in tertiary effluent diluted to 2 percent in reservoir water plus added specific nutrients.															
17		8.6	.019	2	8.6	.048	11		8.7	.005	0	8.7	.012	2	
18	NH_4-N	8.6	.017	4	8.6	.050	7	18.4	8.8	.002	1	8.6	.002	2	3.1
19		8.6	.016	3	8.5	.045	8		8.7	.002	-5	8.6	.002	1	
20		8.6	.010	3	8.6	.070	11		8.8	.001	-2	8.7	.010	6	
21	PO_4-P	8.6	.020	4	8.5	.080	10	32.4	8.8	.002	-3	8.6	.020	7	7.2
22		8.5	.040	5	8.5	.090	13		8.8	.002	-2	8.6	.020	4	
23	Trace	8.5	.032	6	8.6	.070	14		8.8	.001	-1	8.6	.015	4	
24	and	8.5	.032	5	8.5	.060	11	27.5	8.7	.001	-3	8.5	.010	3	2.5
25	Iron	8.5	.034	6	8.6	.070	11		8.8	.002	-3	8.6	.005	3	
26		8.8	.062	11	8.5	.120	25		8.8	.012	0	8.6	.025	5	
27	NAAM	8.8	.063	11	8.6	.140	28	47.8	8.9	.010	-3	8.6	.025	7	10.6
28		8.8	.068	8	8.6	.140	26		8.9	.012	-2	8.6	.020	6	
M. Add Back studies: Selenastrum capricornutum growth in test secondary effluent diluted to 2 percent in reservoir water plus added specific wash products.															
1A									8.8	.005	-2	8.8	.100	18	
1B	+BOLD								8.8	.002	-6	8.8	.105	17	54.6
1C									8.8	.005	-6	8.7	.125	16	
2A									8.8	.005	-3	8.6	.090	14	
2B	+DUZ								8.8	.007	-3	8.7	.130	14	41.9
2C	SOAP								8.9	.005	-6	8.6	.120	16	
3A									8.8	.008	-4	8.8	.120	18	
3B	+SEARS								8.8	.010	-4	8.6	.110	18	45.5
3C									8.9	.010	-3	8.6	.095	12	
4A									8.8	.010	0	8.5	.085	10	
4B	+ECOLO G								8.9	.012	-1	8.6	.120	16	44.0
4C									8.9	.015	-3	8.6	.095	12	

*Biomass measurements are defined as follows: O.D. is the optical density for a one-inch path length cell in a B+L spec. 20 at 750 mμ, PC is particulate carbon in mg C/l, and SS is suspended solids (103°C) for a composite of the three replicate flasks.

**Unusual sample excluded from composite.

Appendix C-2

Maximum Growth Response (18 or 21 Days of Growth) as Particulate Carbon to Dilution Waters Alone

	PC, mg C/l			
	Baseline	Test	Repeat Assay	Random Samples
Selenastrum capricornutum				
Deionized Water	2	0	3	0
Reservoir Water	11	1	1	10.3
Anabaena flos-aquae				
Deionized Water	0	1*	3	0
Reservoir Water	6	2*	3	9.7

	Spiking Study of Repeat Assay Dilution Waters PC, mg C/l			
	+563 µg N/l	+25 µg P/l	+Fe & TE	+13.4% NAAM
Selenastrum capricornutum				
Deionized Water	3.3	5.7	1.7	12.3
Reservoir Water	6.7	3.3	5.3	12.7

*Estimated from OD measurements.

Appendix C-3

Quantities of Materials Added to Two Percent Samples in Reservoir Water

A. Spiking Study

Element	Compound	Percent of Element in NAAM[a]	Concentration in Bioassay Flask µg element/l
N	NH_4Cl	13.4	563
P	KH_2PO_4	13.4	25
Fe	Fe and Trace Elements	13.4	4.4
NAAM	NAAM	13.4	--

B. Add Back Study

Wash Product	mg P/g Product	mg Product/l[b]	µg P/l Addition to Sample
Bold	55.0	0.578	32.0
Duz Soap	0.014	0.578	0.008
Ecolo G	0.0065	0.670	0.004
Sears	0.018	1.172	0.02

[a] NAAM medium is described in Appendix C-4.

[b] Calculated so that the addition of Bold approximately doubled the total phosphorus from the secondary effluent sample. Others were added in direct ratio to Bold according to manufacturers use recommendation on the box of product.

Appendix C-4

Stock Nutrient Solution (PAAP, 1970)

Stock Bottle	Compound	Concentration (mg/l)	Element	Concentration (mg/l)
A_1	$NaNO_3$	25.500	N	4.200
B	K_2HPO_4	1.044	P	0.186
A_2	$MgCl_2$	5.700	Mg	2.904
A_2	$MgSO_4 \cdot 7H_2O$	14.700	S	1.911
A_4	$CaCl_2 \cdot 2H_2O$	4.410	C	2.143
A_3	$NaHCO_3$	15.000	Ca	1.202
			Na	10.999
			K	0.469
C	H_3BO_3	185.640	B	32.450
	$MnCl_2$	265.270	Mn	115.790
	$ZnCl_2$	32.700	Zn	15.683
	$CoCl_2$	0.780	Co	0.354
	$CuCl_2$	0.009	Cu	0.004
	$Na_2MoO_4 \cdot 2H_2O$	7.260	Mo	2.878
D	$FeCl_3$	96.000	Fe	33.043
	$Na_2EDTA \cdot 2H_2O$	300.000		