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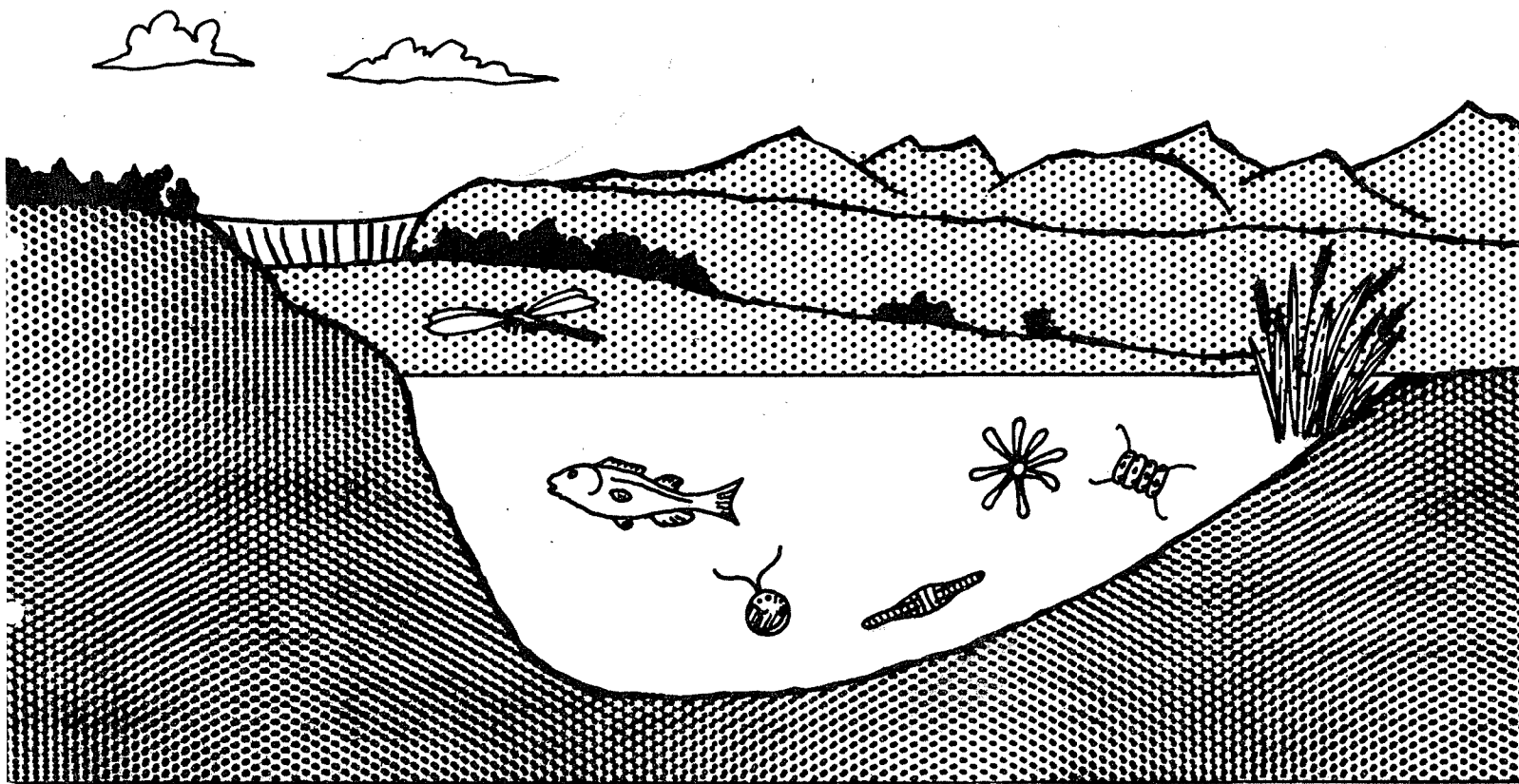
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ALGAL BIOASSAY STUDY **for the** **ANIMAS - LA PLATA PROJECT**

33
By Leslie G. Terry and V. Dean Adams



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Logan, Utah 84321

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LAPLATA PROJECT

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This report was completed for the United States Bureau of Reclamation as part of Contract No. 7-07-40-S0329 (Chemical and Biological Analysis of Colorado Water Samples).

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LIMITING NUTRIENT BIOASSAYS

Sample Pretreatment:

The pretreatment procedure was the same for each of the five sample dates (Sept. 8, 1977; Nov. 29, 1977; Jan. 9, 1978; March 8, 1978; May 10, 1978). Immediately on arrival three liters of each sample was filter sterilized using 0.45 μ Millipore filters. Filtering removes native algae from the test water and enables the use of unialgal test species in the bioassay. Following filtration, the samples were subjected to routine chemical analyses for the determination of indigenous levels of soluble total and ortho phosphorus and soluble total inorganic nitrogen (Table III-1).

Chemical analysis is useful for identifying specific ions but cannot distinguish between biologically available ions and those which are not available. This is where the value of the bioassay lies. Bioassays use the measurable response of living organisms to environmental variables including determining whether or not nutrients are biologically available.

Experiment Set-Up Procedure:

The bioassays were conducted using 100 ml. sample volumes in 500 ml. Erlenmeyer flasks. Inverted beakers were chosen for flask closures in order to permit good CO_2 - O_2 exchange and to prevent contamination.

Prior to use in the bioassays all glass and labware contacting the samples were treated in the following manner: sodium bicarbonate wash, tap water rinses, 1:1 hydrochloric acid rinses, deionized water rinses and finally ultra pure dionized water rinses. Following washing, all glassware was autoclaved using an aluminum foil closure at 121°C for 15 minutes.

Table III-1.

Animas-La Plata Project
Results of Chemical Analyses

	Orthophosphate ($\text{PO}_4\text{-P}$) ($\mu\text{g/l}$)	Total Soluble Phosphorus ($\mu\text{g/l}$)	Ammonia ($\text{NH}_3\text{-N}$) ($\mu\text{g/l}$)	Nitrate+Nitrite ($\text{NO}_3\text{+NO}_2\text{-N}$) ($\mu\text{g/l}$)	Nitrogen/Phosphorus* ($\text{NH}_3\text{+NO}_3\text{+NO}_2\text{-N/PO}_4\text{-P}$)
<u>9/8/1977</u>					
Animas River at Durango	26.	51.	56.	122.	7.
Animas River at 32nd St. Bridge	16.	16.	50.	137.	12.
<u>11/29/1977</u>					
Animas River at Durango	<1.	28.	26.	190.	>216.
Animas River at 32nd St. Bridge	1.	12.	31.	190.	221.
La Plata River near Colo./N.M. Border	1.	10.	8.	370.	378.
<u>1/9/1978</u>					
Animas River at Durango	2.	9.	99.	150.	125.
Animas River at 32nd St. Bridge	1.	9.	45.	150.	195.
La Plata River near Colo./N.M. Border	2.	13.	66.	630.	348.
<u>3/8/1978</u>					
Animas River at Durango	2.	9.	48.	182.	115.
Animas River at 32nd St. Bridge	2.	30.	44.	154.	99.
La Plata River near Colo./N.M. Border	3.	69.	34.	530.	188.
<u>5/10/1978</u>					
Animas River at Durango	1.	1.	24.	100.	124.
Animas River at 32nd St. Bridge	1.	4.	74.	110.	184.
La Plata River near Colo./N.M. Border	19.	22.	54.	360.	22.

*A nitrogen/phosphorus ratio of <15 indicates nitrogen limitation while an N/P ratio of >15 indicates phosphorus limitation.

Samples from the Animas - La Plata project received the treatments listed on Table III-2 for each of the five sampling dates. Each treatment was set up in triplicate. The samples blanks (treatment A and H) were included to provide the basis for comparison of the other treatments and provide a measure of the general fertility of the sample. The control treatments were included to provide an estimate of theoretical cell growth and an index for comparing growth levels in the test waters.

Table III-3 lists the constituents of Algal Assay Medium (AAM). AAM is a precisely prepared growth medium containing known concentrations of all compounds essential to algal growth. The samples and control (with the exception of 9/8/77 controls) contained one half AAM levels of nitrogen and phosphorus whereas all other constituent were added at full strength levels. Di-sodium EDTA (Ethylene dinitrilo tetraacetic acid) a commonly used organic chelator, was added to Treatments A-G at a level of 1 mg/l in order to render excess metal ions biologically inactive. Serious metal toxicity was detected in earlier bioassays from the area. Treatment H and I (without EDTA) were included to confirm the metal toxicity. Increased growth in EDTA spiked samples in comparison to yields in the untreated flasks can be directly attributable to organic chelation and consequently metal toxicity.

Algal bioassays were performed according to EPA (1971) using the Green Alga, Selenastrum capricornutum PRINTZ. The test flasks were placed in a constant temperature room ($24^{\circ} \pm 2^{\circ}\text{C}$) with "cool white" fluorescent lighting providing illumination of 400 ft-C (4304 lux) \pm 10 percent.

The algal assays were monitored by determining the optical density (OD, Bausch and Lomb spectrophotometer to 750 nm, 1 cm path length) and

Table III-3. Algal assay medium (AAM)

Compound		Concentration in NAAM	
		Compound mg/l	Element mg/l
A ₁	NaNO ₃	25.500	N 4.2
A ₂	MgCl ₂ 6H ₂ O	12.171	Mg 2.9
	MgSO ₄ 7H ₂ O	14.700	
A ₃	CaCl ₂ 2H ₂ O	4.410	Ca 1.2
A ₄	NaHCO ₃	15.000	
B	K ₂ HPO ₄	1.044	P 0.186
		<u>µg/l</u>	<u>µg/l</u>
C	H ₃ BO ₃	185.64	B 32.45
	MnCl ₂ 4H ₂ O	417.18	Mn 115.80
	ZnCl ₂	32.70	Zn 15.68
	Na ₃ MoO ₄ 2H ₂ O	7.26	Mo 2.88
	CoCl ₂ 6H ₂ O	1.43	Co 0.35
	CuCl ₂ 2H ₂ O	0.01	Cu 0.004
D	FeCl ₃ 6H ₂ O	160	Fe 33.05
	Na ₂ EDTA 2H ₂ O	300	<u>mg/l</u>
Protocol for Nutrient Spiking			S 1.91
A ₁	Nitrogen		Na 11.04
B	Phosphorus		K 0.47
A ₁ + B	N + P		C 2.14
C + D	Trace Elements (T. E.)		
ALL	NAAM		

Reference: Environmental Protection Agency, "Algal Assay Procedures: Bottle Test". Corvallis, Oregon. (1971) 82 pages.

Table III-2.

Animas - La Plata Project
Treatment Constituents

9/8/77

- A. Sample + 1 mg/l EDTA
- B. Sample + 1 mg/l EDTA + 2.1 mg/l Nitrogen (N)
- C. Sample + 1 mg/l EDTA + 0.09 mg/l Phosphorus (P)
- D. Sample + 1 mg/l EDTA + 2.1 mg/l N + 0.093 mg/l P
- E. Sample + 1 mg/l EDTA + Trace Elements (AAM Levels)
- F. Sample + 1 mg/l EDTA + 15.0 mg/l NaHCO_3
- G. Sample + 1 mg/l EDTA + 2.1 mg/l N + 0.093 mg/l P + AAM levels of:
trace elements, NaHCO_3 , CaCl_2 and MgSO_4
- H. Sample
- I. Sample + 2.1 mg/l N + 0.093 mg/l P + AAM levels of:
trace elements, NaHCO_3 , CaCl_2 and MgSO_4
- Control: Distilled water + 4.2 mg/l N + 0.186 mg/l P + AAM levels of:
trace elements, NaHCO_3 , CaCl_2 and MgSO_4
- Control + EDTA: Distilled water + 1 mg/l EDTA + same as control above

11/29/77

Animas River at Durango
Animas River at 32nd St. Bridge
La Plata River at Colo./N.M. Border

Sample Treatments Same as 9/8/77

Control: Distilled Water + 2.1 mg/l N + 0.093 mg/l P + AAM levels of:
trace elements, NaHCO_3 , CaCl_2 and MgSO_4 .

Control + EDTA: Distilled Water + 1 mg/l EDTA + same as control above

1/9/78

Animas River at Durango
Animas River at 32nd St. Bridge
La Plata River at Colo./N.M. Border

Sample Treatments and Controls same as 11/29/77

3/8/78

Animas River at Durango
Animas River at 32nd St. Bridge
La Plata River at Colo./N.M. Border

Sample Treatments and Controls same as 11/29/77

Table III- 2.
Animas - La Plata Project
Treatment Constituents

5/10/78

Animas River at Durango
Animas River at 32nd St. Bridge

Sample Treatments and Controls same as 11/29/77

La Plata River at Colo./N.M. Border

- A. Sample
- B. Sample + 2.1 mg/l N
- C. Sample + 0.093 mg/l P
- D. Sample + 2.1 mg/l N + 0.093 mg/l P
- E. Sample + trace elements (AAM levels)
- F. Sample + 15.0 mg/l NaHCO_3
- G. Sample + 2.1 mg/l N + 0.093 mg/l P + AAM levels of:
trace elements, NaHCO_3 , CaCl_2 , and MgSO_4

Control: same as 11/29/77

relative fluorescence (RF x 30, Turner Fluorometer, Model 110). Optical density was measured over a 14 day period while relative fluorescence was measured to monitor the progress of the cultures for the first six to seven days when optical density does not provide a great deal of sensitivity. Fluorescence is a physiological response measuring chlorophyll a and optical density is a measurement of biomass. Although they are different measurements, the two can be correlated. Normally when chlorophyll a is increasing so is biomass and vice versa. The results of both determinations are represented graphically in Figure 1-54. Maximum values for optical density are listed on Table III-4.

Optical density (OD) is an indirect means of measuring algal cell biomass. As a consequence OD is linearly related to biomass as dry weight (Porcella et al., 1973). Due to this linearity biomass, as volatile suspended solids (V.S.S.) can be calculated directly from OD. The relationship used to convert OD to V.S.S. in Table III-5 is:

$$\text{V.S.S., mg/l} = 350 (\text{OD}) + 3.5$$

Because of the difficulty of measuring biomass in low density cultures, relative fluorescence of in vivo chlorophyll a was used to estimate biomass in the early phases of the bioassay. Maximum values for relative fluorescence are listed on Table III-6. Calculations of average maximum specific growth rate batch ($\hat{\mu}_b$) were made using relative fluorescence. The maximum specific growth rate occurs during the logarithmic phase of growth; usually between day 0 and day 5. Maximum specific growth rates are calculated on Table III-7.

Table III-4

Animas - La Plata Project

Maximum Amount of Growth Observed as Optical Density; 750 nm., 1 cm.

[illegible]

Table III-5.

Animas - La Plata Project

Maximum Amount of Growth Observed as mg/l VSS.^a

[illegible]

Table III-5.
 Animas - La Plata Project
 Maximum Amount of Growth Observed as mg/l VSS.^a

Sample	Treatment								
	A	B	C	D	E	F	G	H	I
<u>5/10/78</u>									
Animas River at Durango	5.6	5.6	5.3	94.2	6.0	6.0	87.5	8.8	34.7
Animas River at 32nd St. Bridge	6.3	6.0	12.3	103.3	6.7	6.0	110.3	8.8	25.9
La Plata River at Colo./N.M. Border	20.3	31.5	24.9	91.0	27.3	20.7	122.5		
Control							96.0		
Control + EDTA									95.9

^aVSS = Volatile Suspended Solids

VSS, mg/l = 350 (Optical Density) + 3.5 (Porcella, et al., 1973)

Maximum Amount of Growth Observed as Relative Fluorescence, RF x 30

[illegible]

Table III-7.

Animas - La Plata Project

Animas - La Plata Project
Maximum Specific Growth Rate; $\hat{\mu}_b$, days⁻¹ a

[illegible]

Table III-7.

Animas - La Plata Project

Maximum Specific Growth Rate, $\hat{\mu}_b$, days⁻¹ ^a

Sample	Treatment								
	A	B	C	D	E	F	G	H	I
5/10/78									
Animas River at Durango	0.07	0.04	0.13	1.47	0.07	0.09	1.56	0.14	0.97
Animas River at 32nd St. Bridge	0.16	0.10	0.60	1.20	0.16	0.08	1.78	0.04	0.88
La Plata River at Colo./N.M. Border	0.72	0.88	1.01	1-52	0.90	0.71	1.68		
Control							1.25		
Control + EDTA									1.29

^aThe maximum specific growth rate ($\hat{\mu}_b$) for an individual treatment is the largest specific growth rate (μ_b) occurring at any time during incubation. The specific growth rate, μ_b , is defined by:

$$\mu_b = \frac{\ln(X_2/X_1)}{t_2 - t_1} \text{ days}^{-1}$$

where

X_2 = biomass concentration at end of selected time interval

X_1 = biomass concentration at beginning of selected time interval

$t_2 - t_1$ = elapsed time (in days) between selected determinations of biomass

Results and Interpretation:

Table III-8 indicates the nutrient limitation as a result of chemical analysis and as a result of algal bioassay for all sites on the Animas-La Plata Project. While nitrogen and phosphorus are most often the algal growth limiting nutrients, it should be recognized that other nutrients may be growth limiting. Theoretical productivity potential must be verified by actual algal assay analyses to determine: (1) the presence of growth limiting nutrients; (2) the presence of toxicants such as heavy metals and (3) if the chemical analyses for N and P is realistic.

1. Animas River at Durango - Figures 1-20

In September 1977 this river sample was phosphorus limited first and then nitrogen limited. The following bioassays in November, 1977; January 1978; March, 1978; and May, 1978 showed similar responses. Without the addition of nutrients, there was only limited response, if any at all, and the river seemed to be oligotrophic. However, the most important fact determined as a result of algal assay was a severe metal toxicity problem on the Animas River at Durango. Growth without addition of Ethylenedinitrite tetraacetic acid (EDTA) ranged from approximately 10 percent to 40 percent of normal depending on the heavy metal concentration during any particular sampling period. Organic chelation of heavy metals with EDTA created conditions more conducive to algal growth and as a result growth was 80 to 100 percent of normal as compared to an AAM control.

Based on a limited amount of heavy metal data available for the Animas River at Durango, it appears likely that zinc was the toxic

Table III-8.

Animas - La Plata Project

Limiting Nutrients

Sample	Limiting Nutrient(s)	
	Chemical Analysis	Bioassay
<u>9/8/77</u>		
Animas River at Durango	Nitrogen	Phosphorus ¹
Animas River at 32nd St. Bridge	Nitrogen	Phosphorus ¹
<u>11/29/77</u>		
Animas River at Durnago	Phosphorus	Nitrogen & Phosphorus
Animas River at 32nd St. Bridge	Phosphorus	Nitrogen & Phosphorus
La Plata River at Colo./N.M. Border	Phosphorus	Phosphorus ¹
<u>1/9/78</u>		
Animas River at Durango	Phosphorus	Phosphorus ¹
Animas River at 32nd St. Bridge	Phosphorus	Phosphorus ¹
La Plata River at Colo./N.M. Border	Phosphorus	Phosphorus ¹
<u>3/8/78</u>		
Animas River at Durango	Phosphorus	Phosphorus ¹
Animas River at 32nd St. Bridge	Phosphorus	Phosphorus ¹
La Plata River at Colo./N.M. Border	Phosphorus	Phosphorus ¹
<u>5/10/78</u>		
Animas River at Durango	Phosphorus	Nitrogen & Phosphorus
Animas River at 32nd St. Bridge	Phosphorus	Nitrogen & Phosphorus
La Plata River at Colo./N.M. Border	Phosphorus	Nitrogen & Phosphorus

¹ Addition of phosphorus substantially increases the maximum specific growth rate; $\hat{\mu}_b$ (Table 7) indicating phosphorus limitation. However, due to the low level of both indigenous nitrogen and phosphorus growth is only minimal upon phosphorus addition (Table 5) as nitrogen becomes limiting as well.

metal in the river. Studies conducted by Joseph C. Green, et al. (1976) on algal growth in Long Lake, Washington have shown that zinc, cadmium or copper create toxic conditions for algae. Neither copper nor cadmium were at toxic concentrations during any of the five sampling dates. In fact both of these elements remained at a level below the detectable limit of the Varian Atomic Absorption Spectrophotometer used to measure the elements. On the other hand a definite correlation between zinc concentration and algal biomass was noted as represented by Figure 1-A. Greene, et al., found that zinc levels of $0.003 - 0.121 \text{ mg/l}^{-1}$ were toxic on Long Lake. The zinc levels on the Animas River at Durango were within this range and higher.

The zinc concentration remained high throughout the fall and winter months but the last assay in May, 1978 indicated a dilution effect on the zinc concentration probably due to the spring turnover. Biomass in Treatment H increased from an average of 11.5 mg/l V.S.S. to 34.7 mg/l V.S.S. Nevertheless, the zinc concentration was still high enough to limit increase in biomass upon N and P addition to less than 40 percent of normal.

As a result of the high zinc concentration in the Animas River at Durango, it seems unlikely that productivity will increase even upon addition of higher concentrations of nitrogen and phosphorus. The main concern appears to be the heavy metals concentration and consequently the possible toxic effect on aquatic organisms. Chapter IV provides a more thorough discussion of the trophic status on the Animas River.

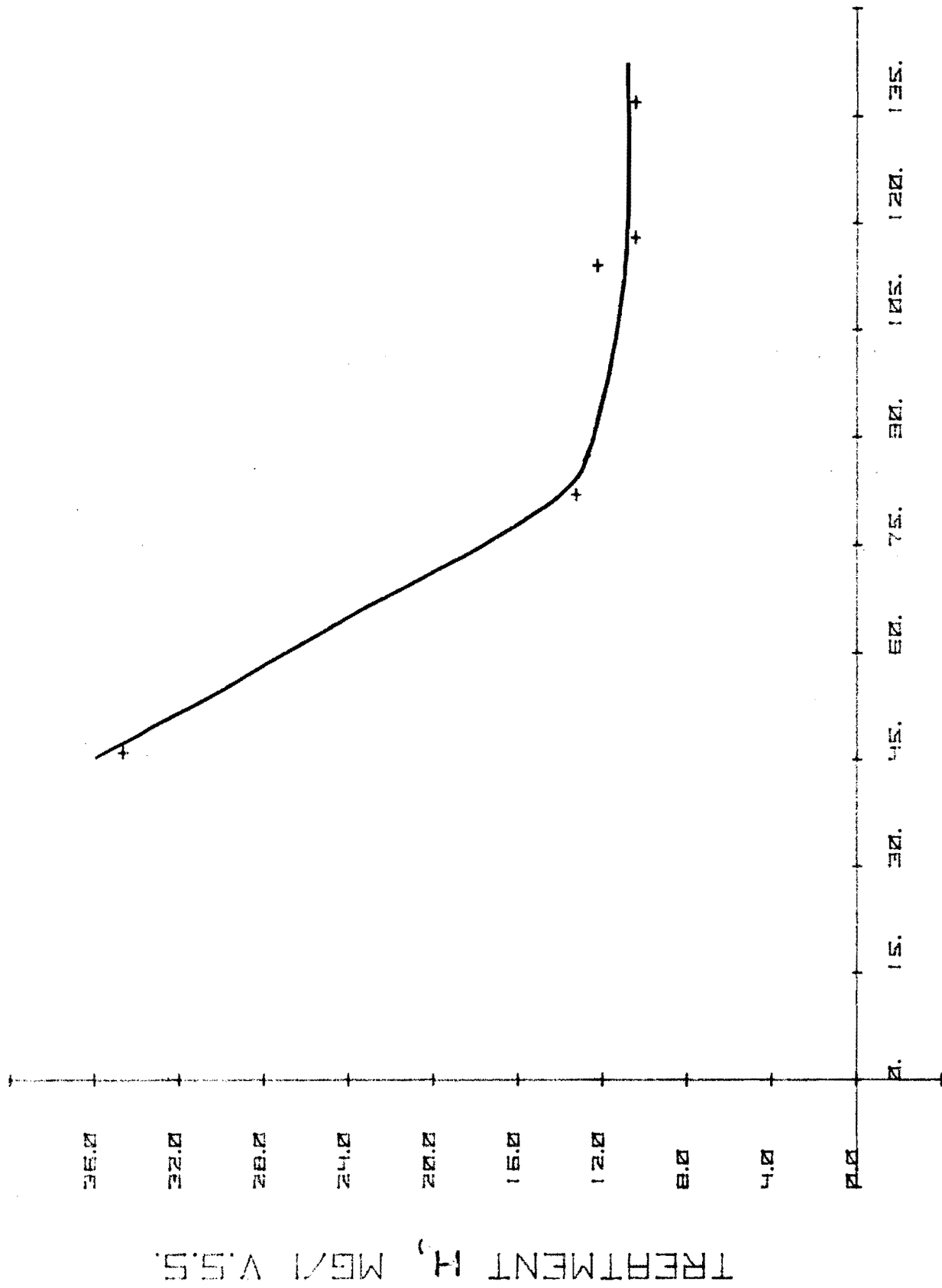


Figure 1-A. ZINC CONCENTRATION, UG/L

ANIMAS RIVER AT DURANGO
SEPTEMBER 8 1977

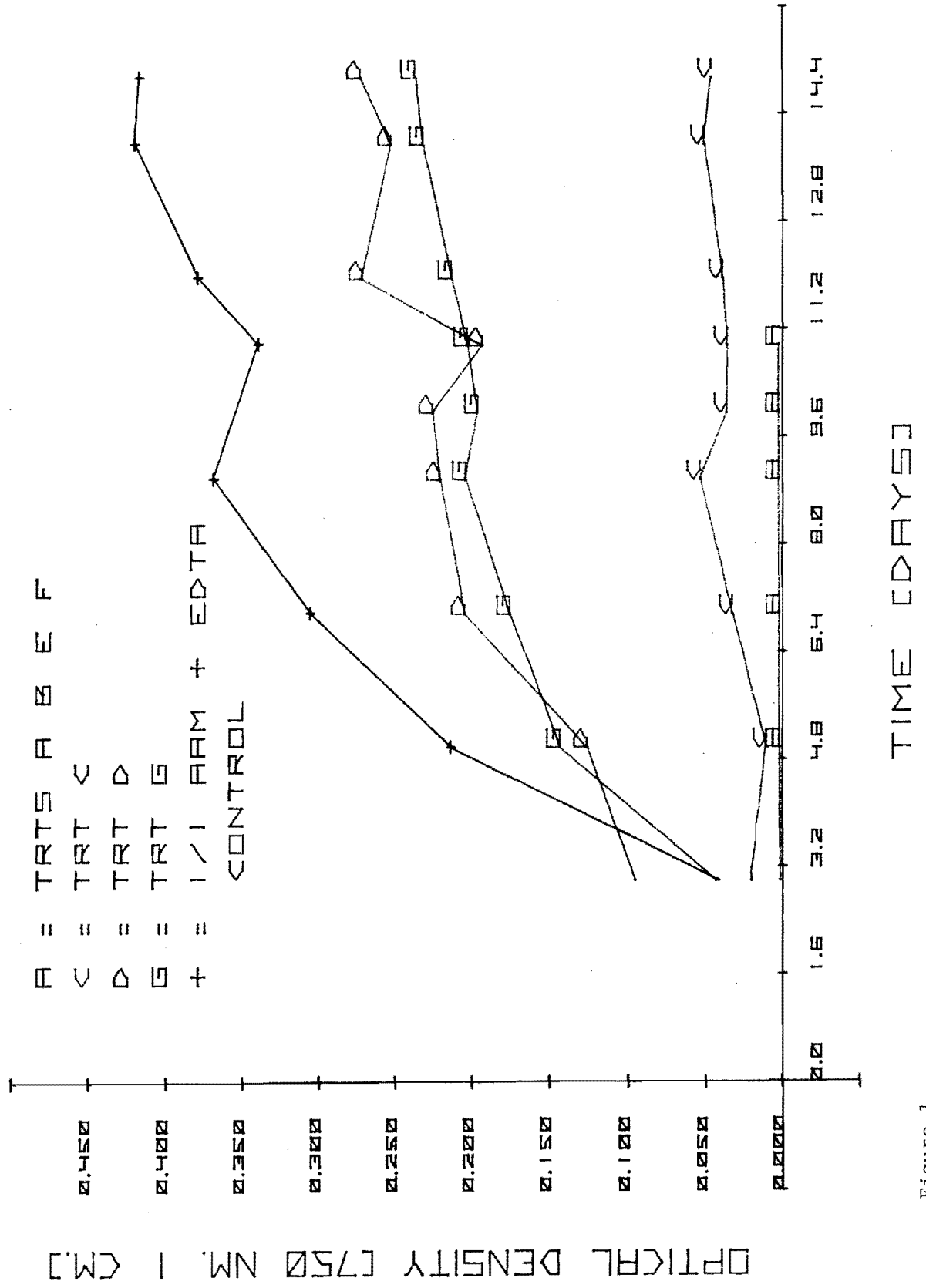


Figure 1.

ANIMAS RIVER AT DURANGO
SEPTEMBER 8 1977

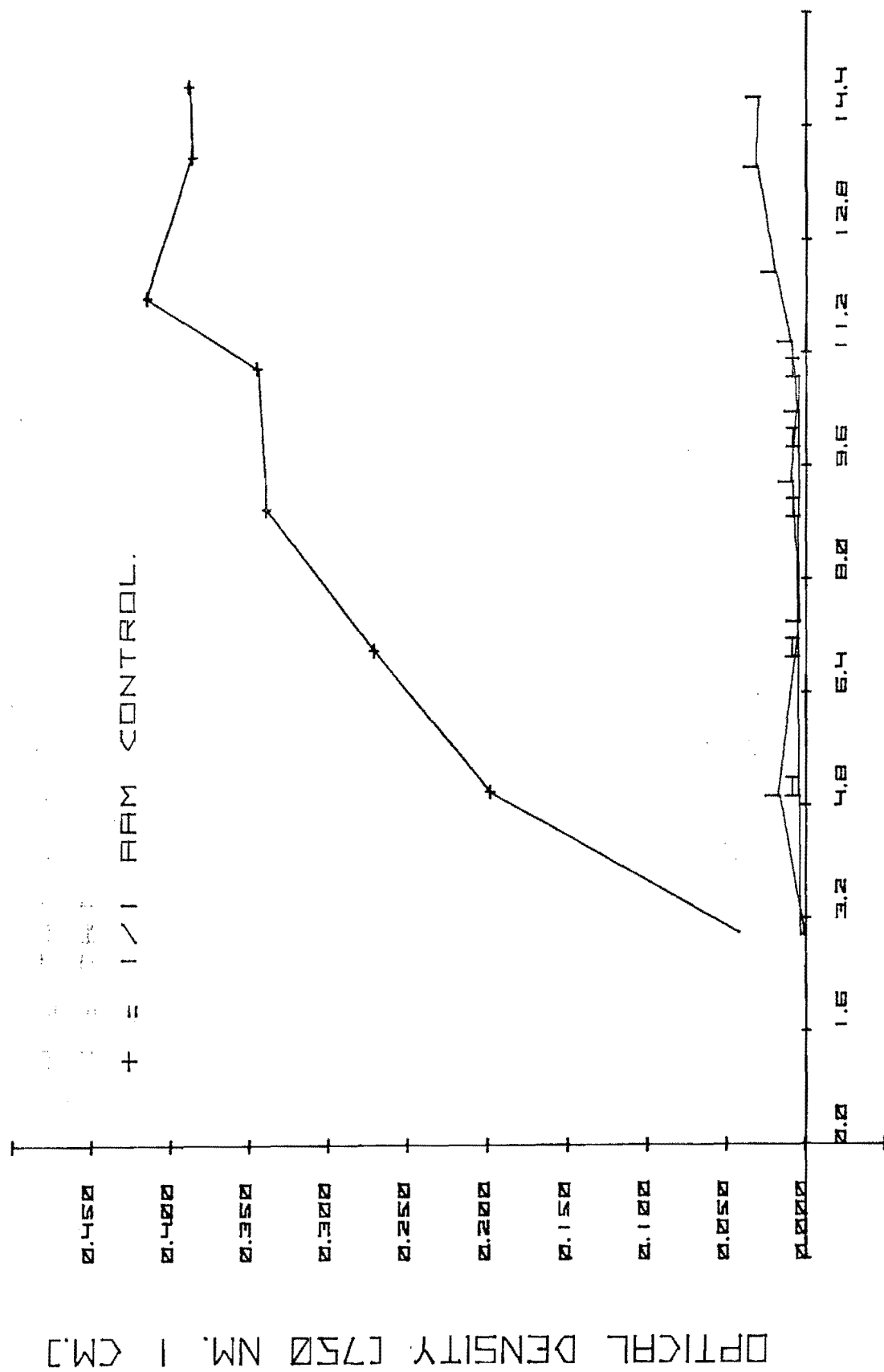


Figure 2.

TIME (HOURS)

ANIMAS RIVER AT DURANGO
SEPTEMBER. 8 1977

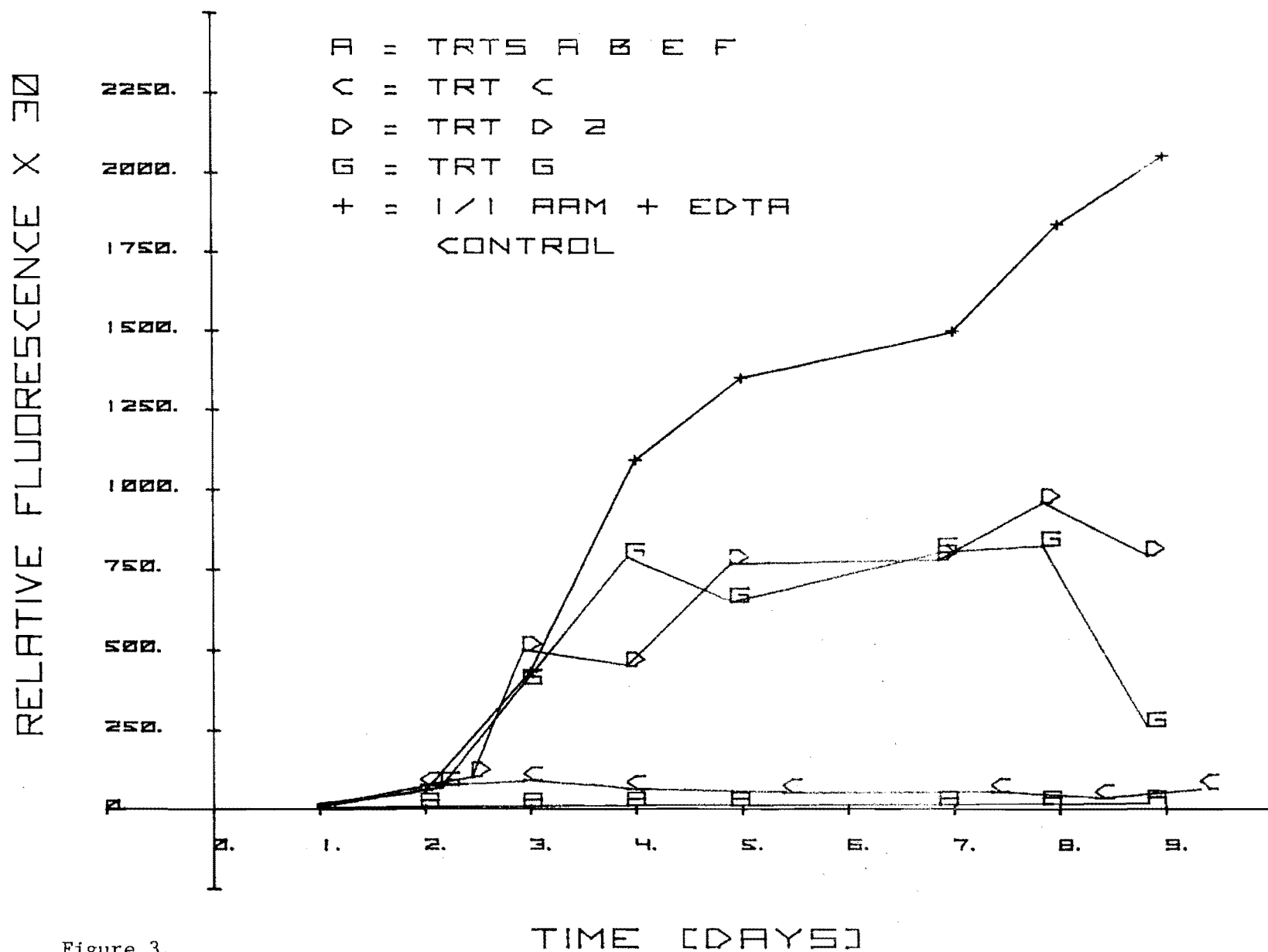


Figure 3.

ANIMAS RIVER AT DURANGO

SEPTEMBER 8 1977

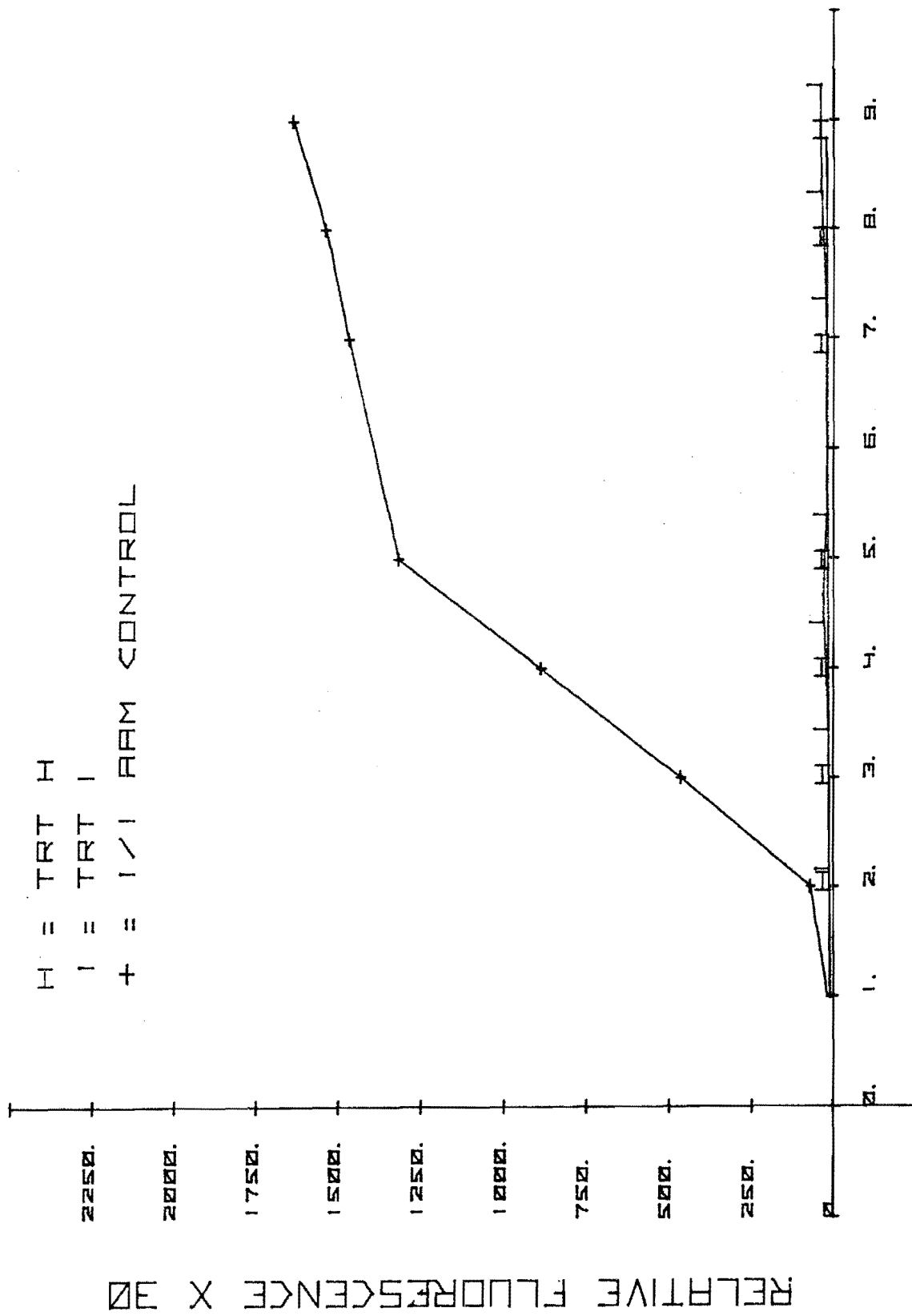


Figure 4.

ANIMAS RIVER AT DURANGO
NOVEMBER 29 1977

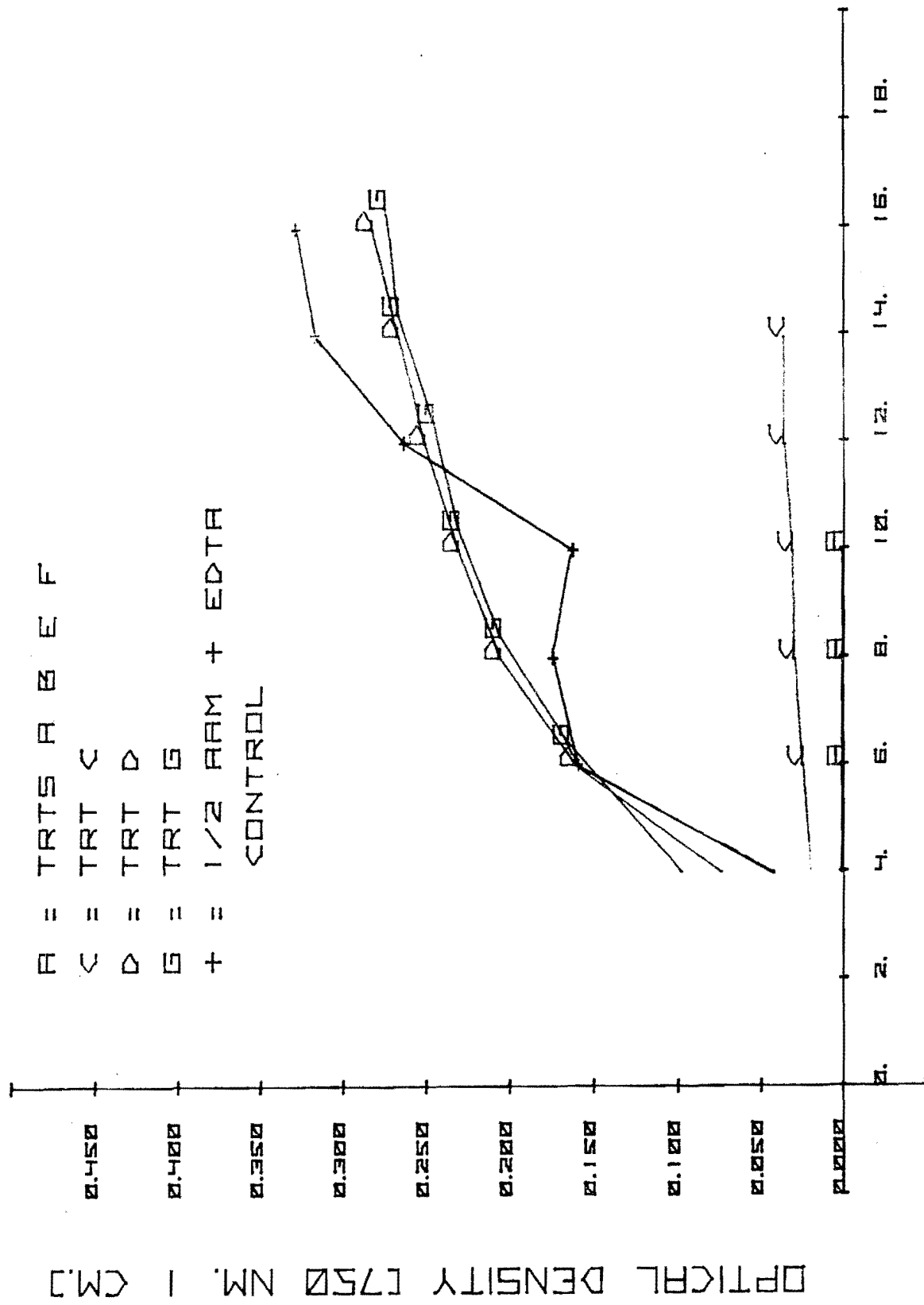


Figure 5.

TIME [DAYS]

ANIMAS RIVER AT DURANGO
NOVEMBER 29 1977.

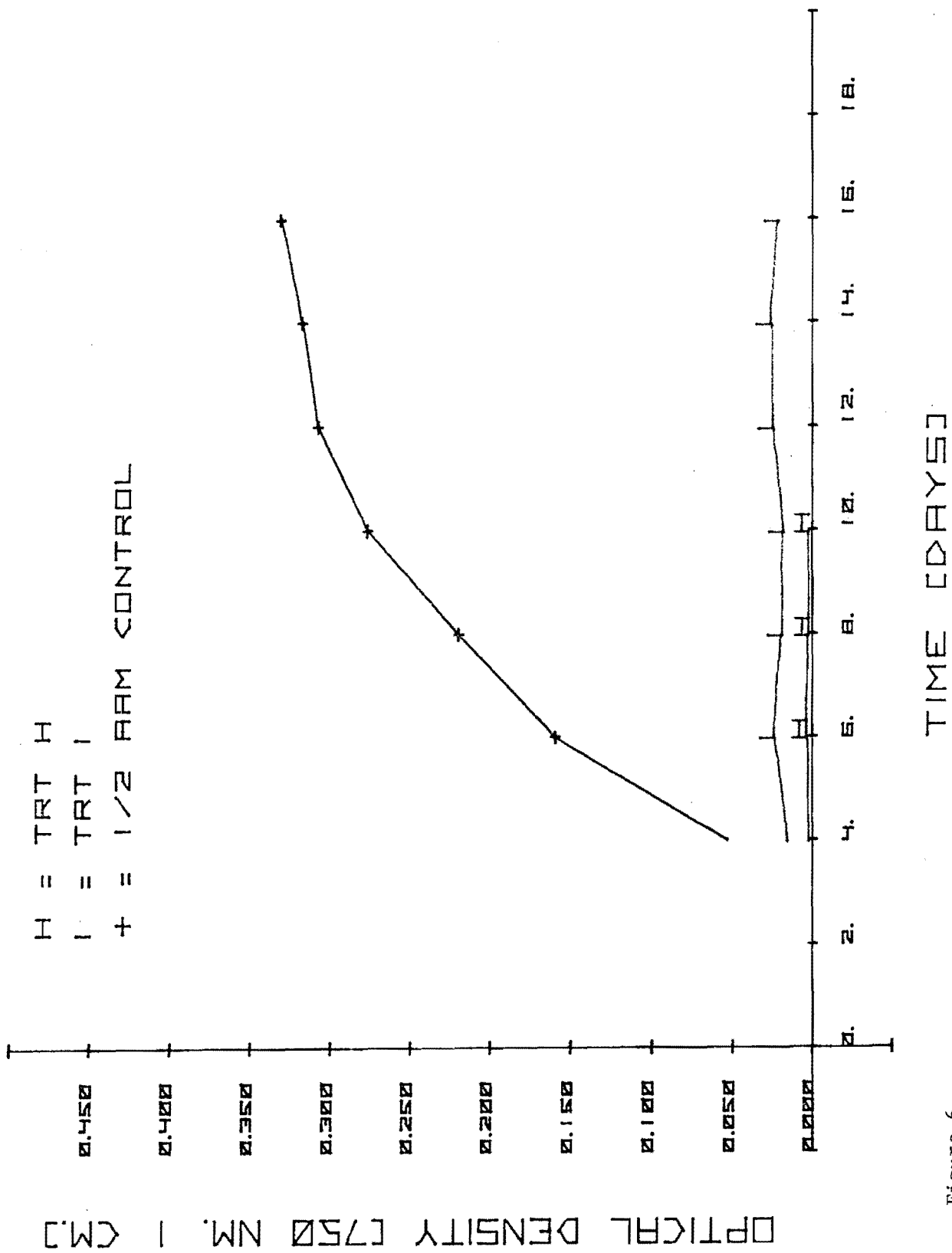


Figure 6.

ANIMAS RIVER AT DURANGO

NOVEMBER 29 1977

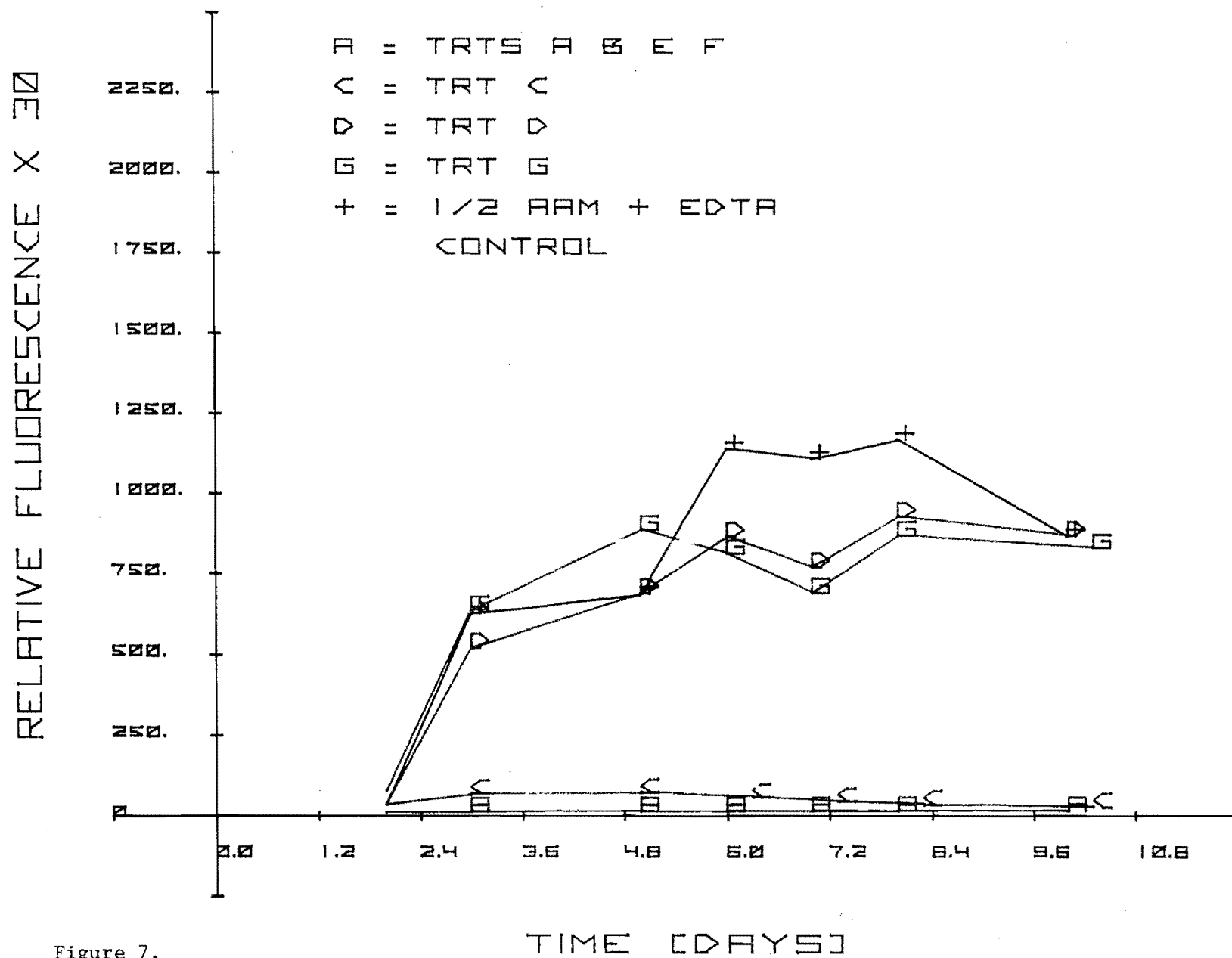


Figure 7.

ANIMAS RIVER AT DURANGO
 NOVEMBER 29, 1977.

H = TRT H
 I = TRT I
 + = 1/2 ARM CONTROL

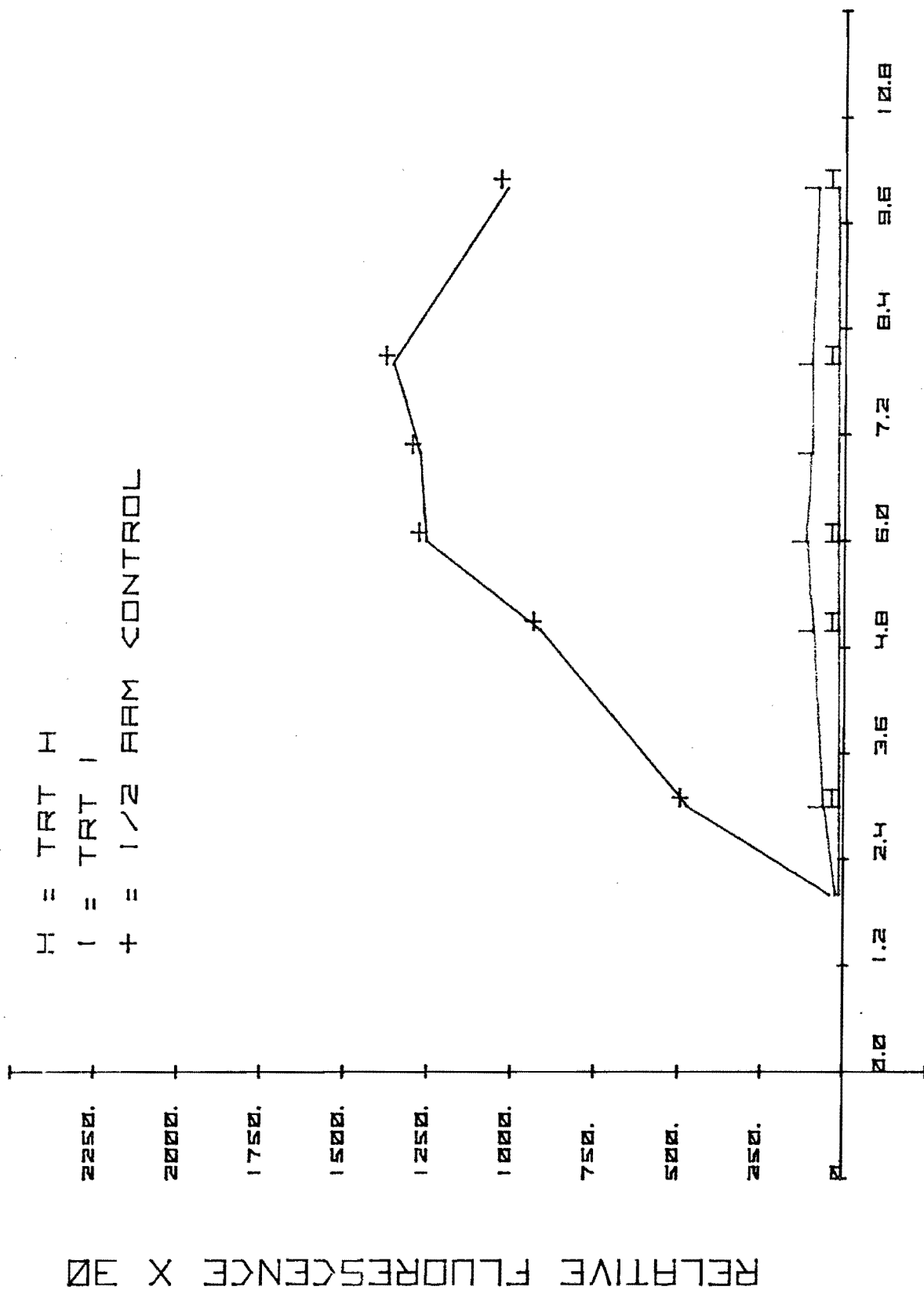


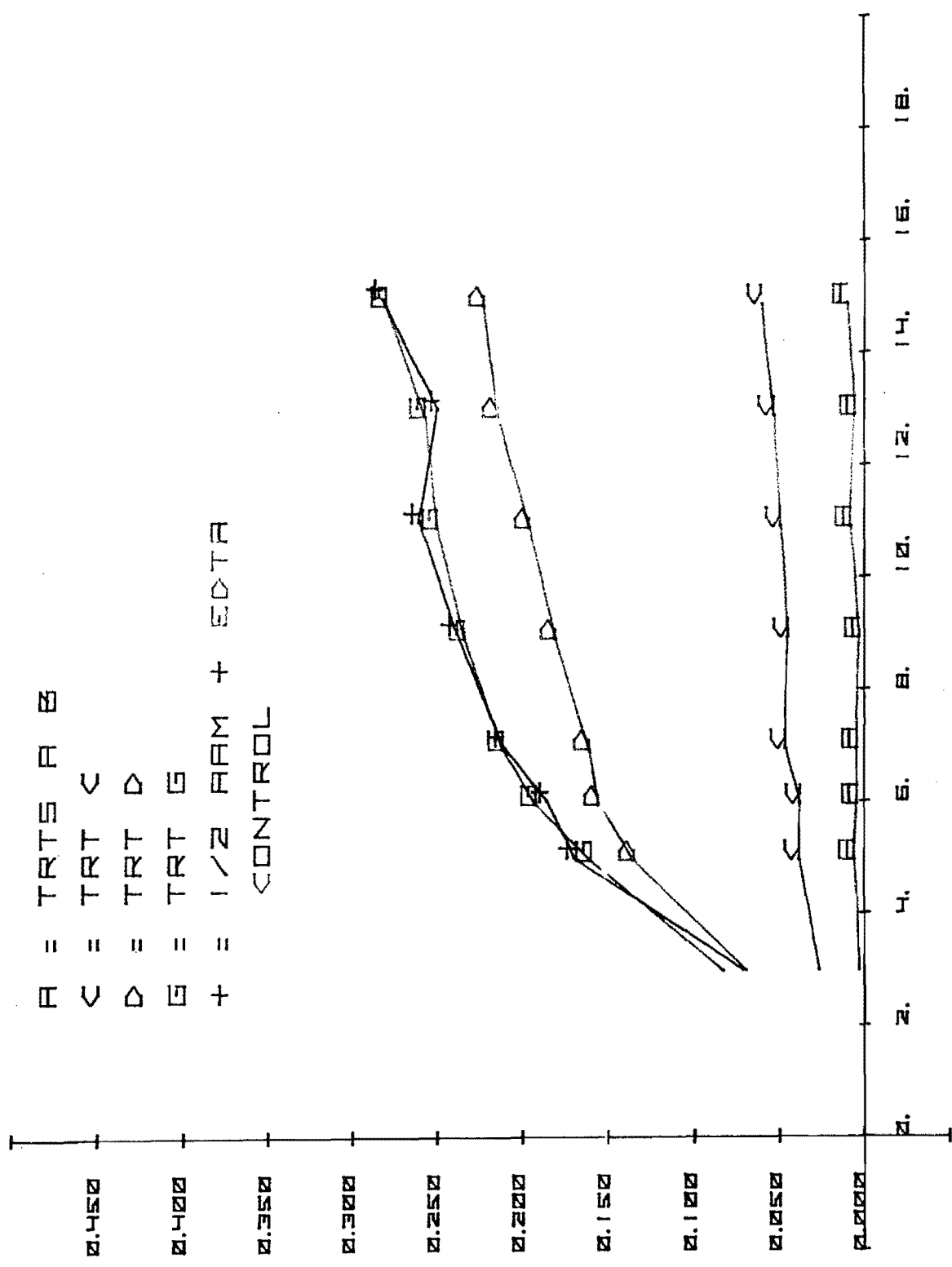
Figure 8.

ANIMAS RIVER AT DURANGO

JANUARY 9 1978

A = TRTS A B
 C = TRT C
 D = TRT D
 G = TRT G
 + = 1/2 ARM + EDTA
 CONTROL

OPTICAL DENSITY [750 NM. 1 CM.]



TIME [DAYS]

Figure 9.

ANIMAS RIVER AT DURANGO
JANUARY 9 1978

H = TRT H
I = TRT I
+ = 1/2 AM CONTROL

OPTICAL DENSITY [750 NM. 1 CM.]

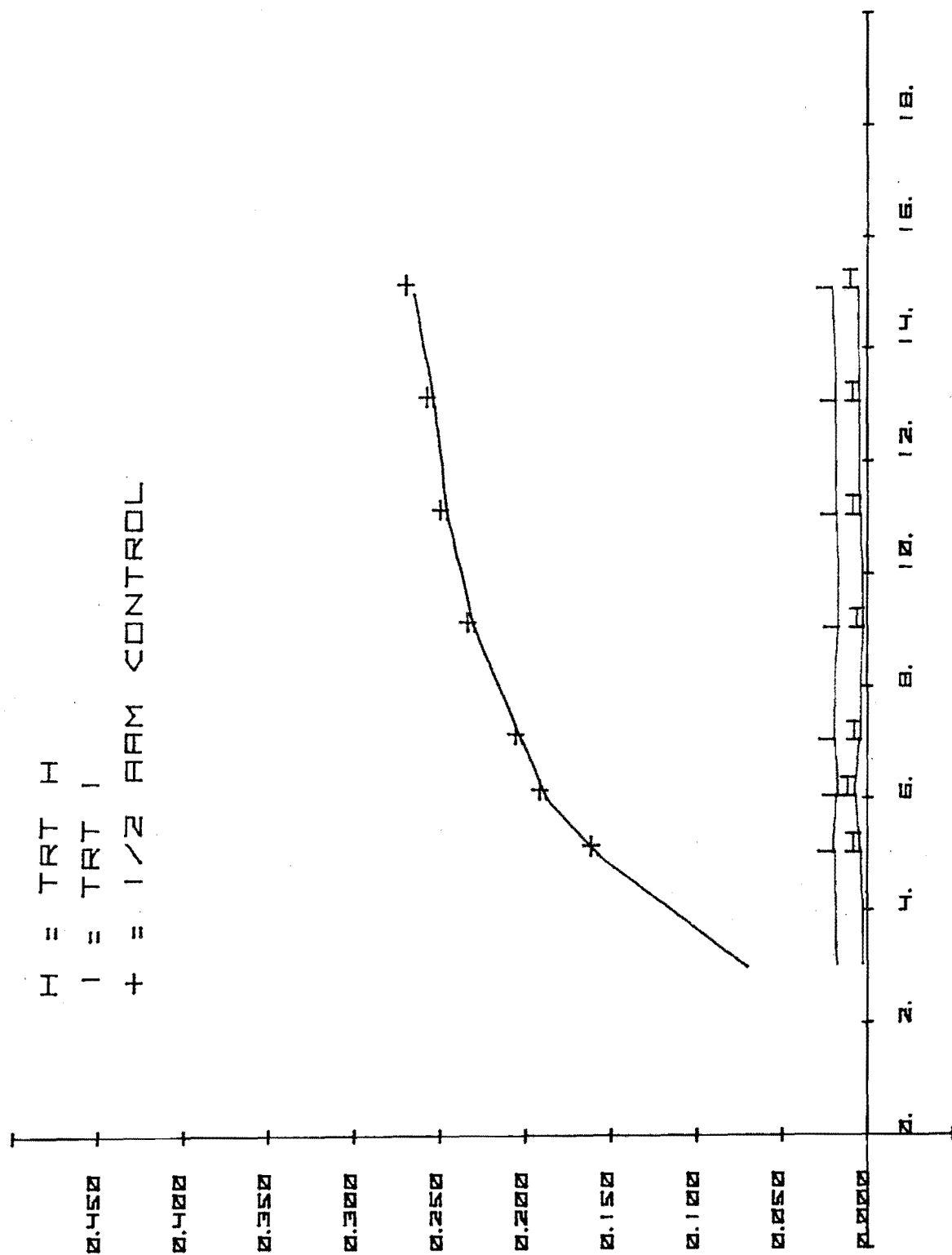


Figure 10.

ANIMAS RIVER AT DURANGO
JANUARY 9 1978

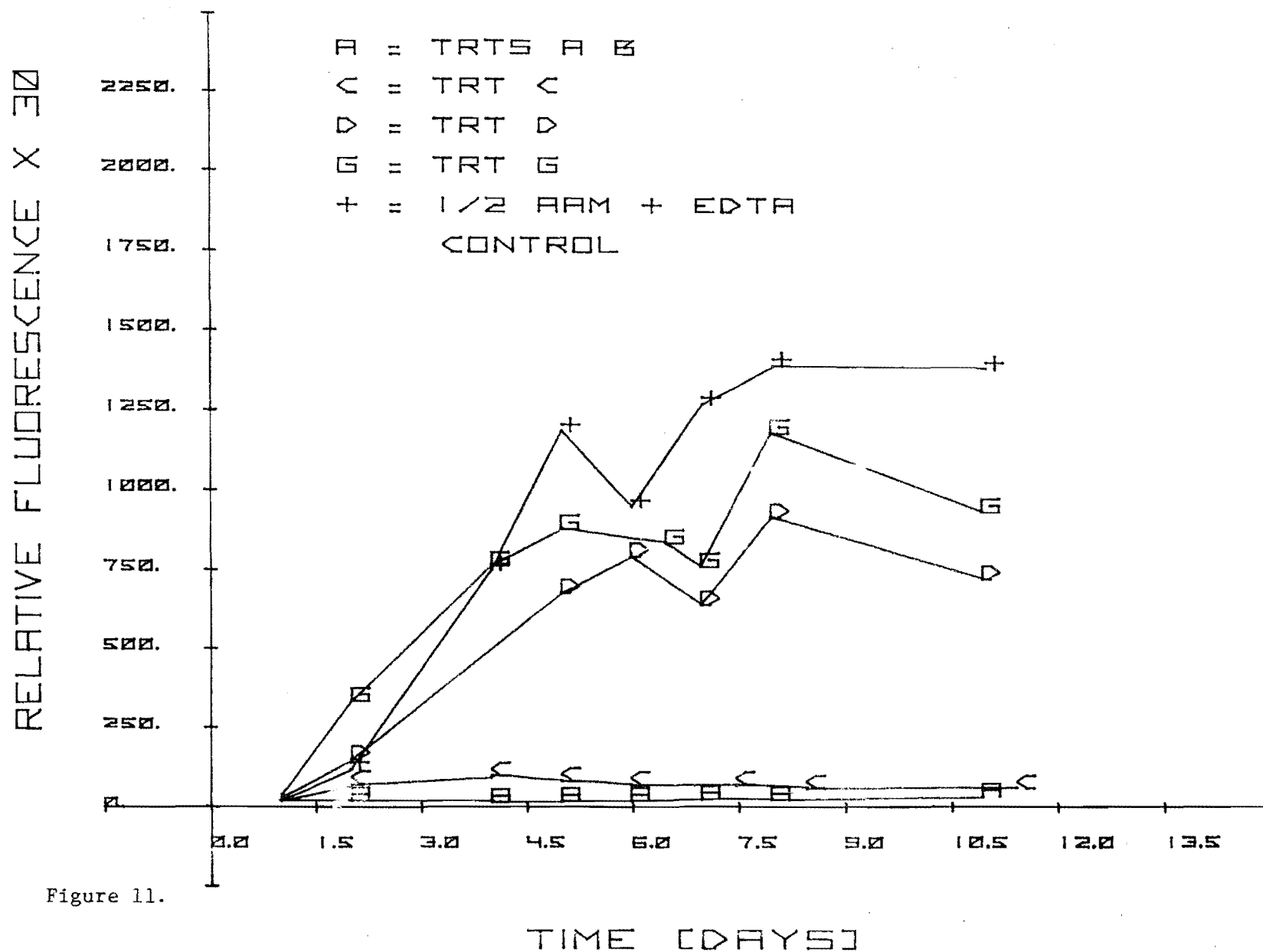


Figure 11.

ANIMAS RIVER AT DURANGO
JANUARY 9 1978

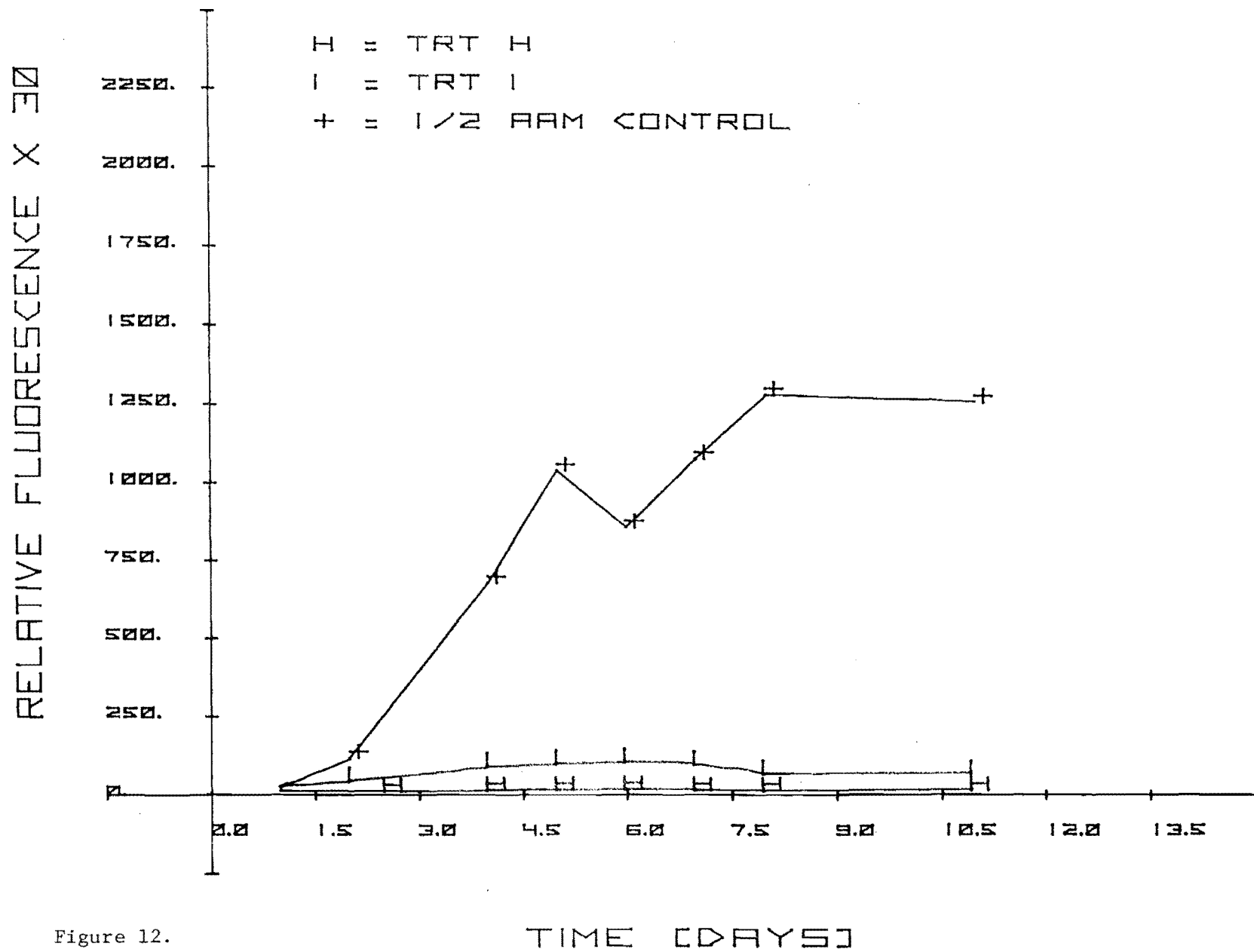


Figure 12.

ANIMAS RIVER AT DURANGO
MARCH 8 1978

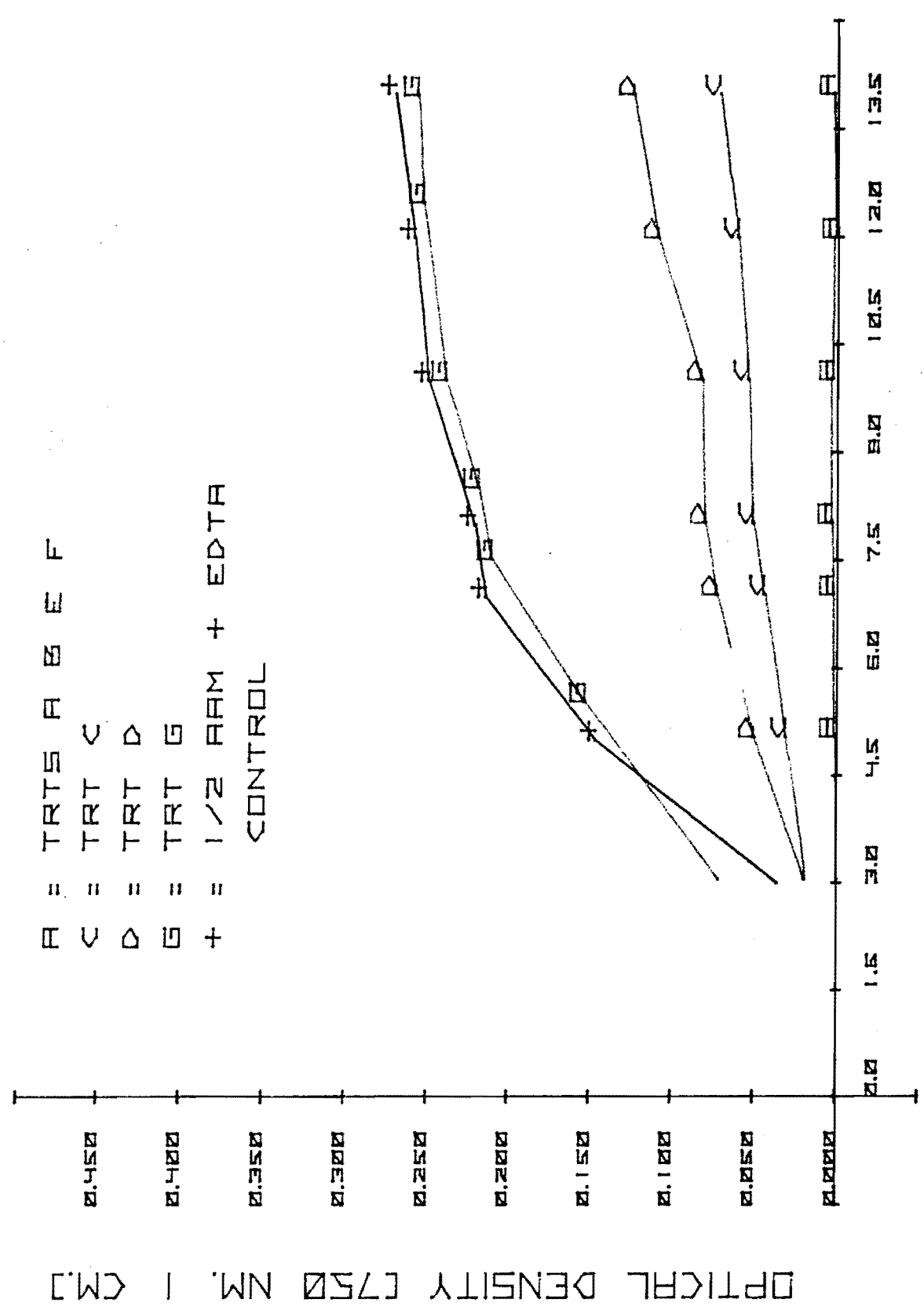


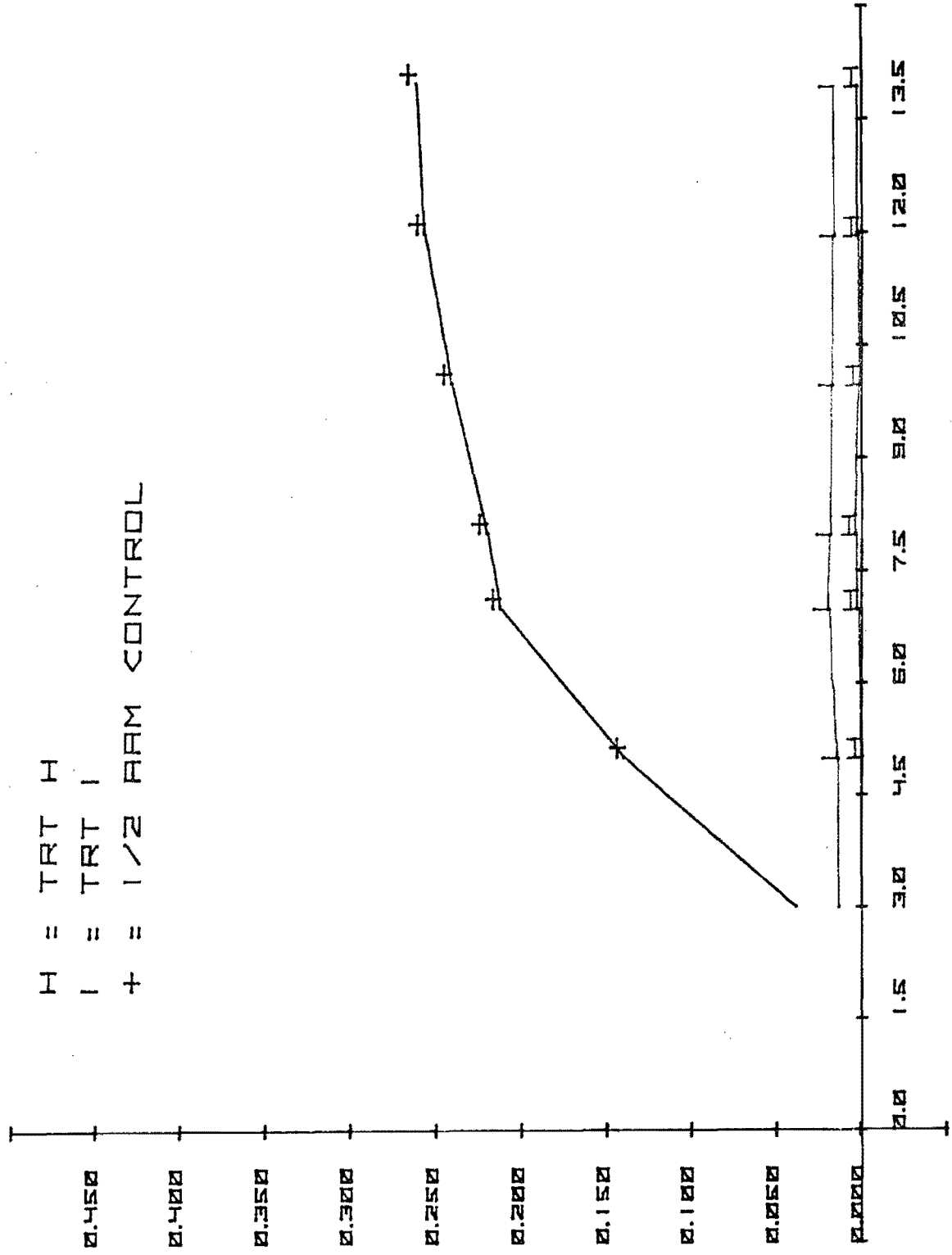
Figure 13.

TIME [DAYS]

ANIMAS RIVER AT DURANGO
MARCH 8 1978

H = TRT H
I = TRT I
+ = 1/2 ARM CONTROL

OPTICAL DENSITY [750 NM. 1 CM.]



TIME (DAYS)

Figure 14.

ANIMAS RIVER AT DURANGO
MARCH 8 1978

RELATIVE FLUORESCENCE X 30

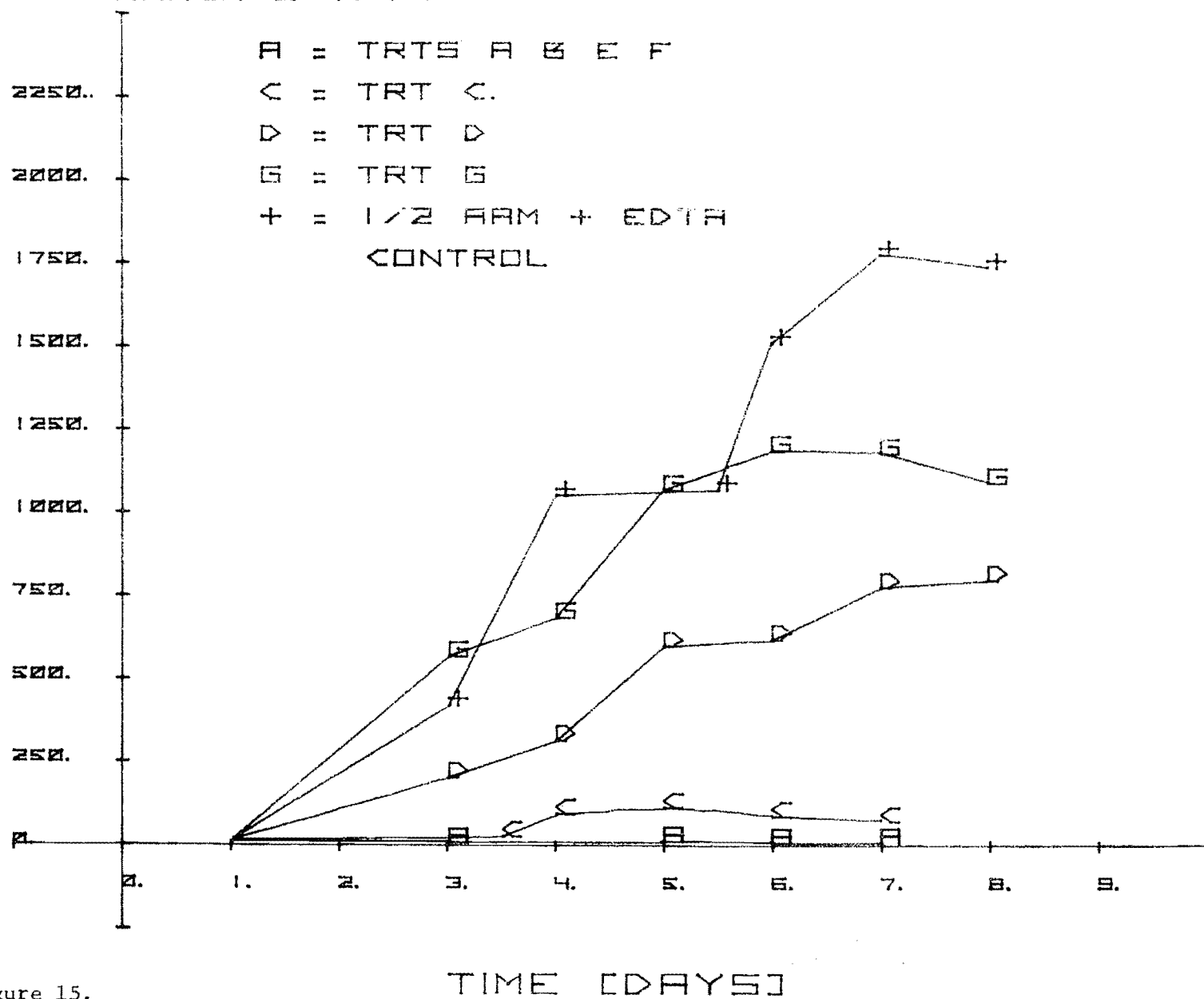


Figure 15.

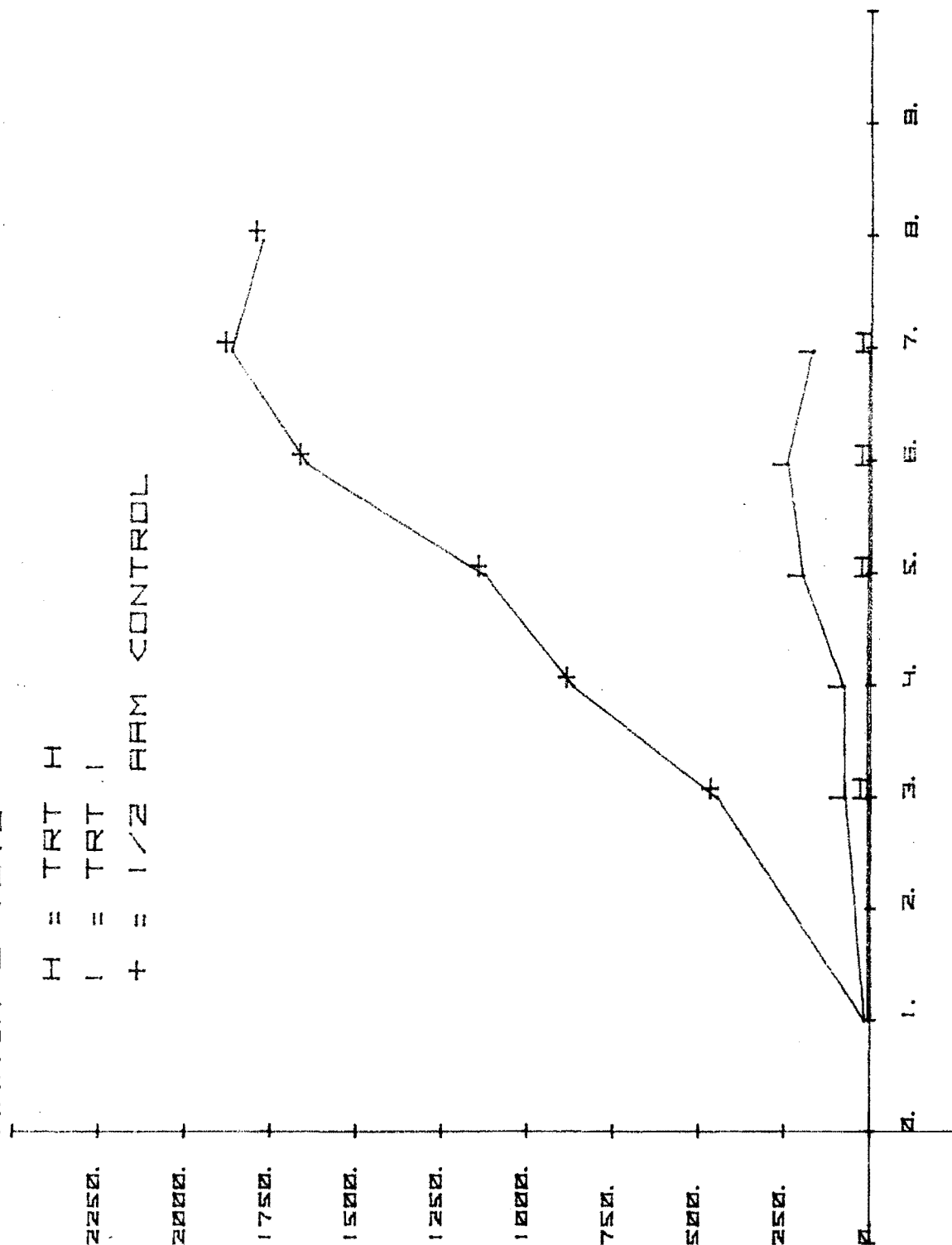
ANIMAS RIVER AT DURANGO
MARCH 8 1978

H = TRT H

I = TRT I

+ = 1/2 HAM CONTROL

RELATIVE FLUORESCENCE X 10³



TIME [DAYS]

Figure 16.

OPTICAL DENSITY, [750 NM. 1 CM.]

ENRICHED RIVER IT OILHEMED
MAY 12, 1972

D = TRT E, B, C, E, F + EDTH
 Δ = TRT Δ + EDTH
 E = TRT E + EDTH
 + = 1/2 TRM CONTROL + EDTH

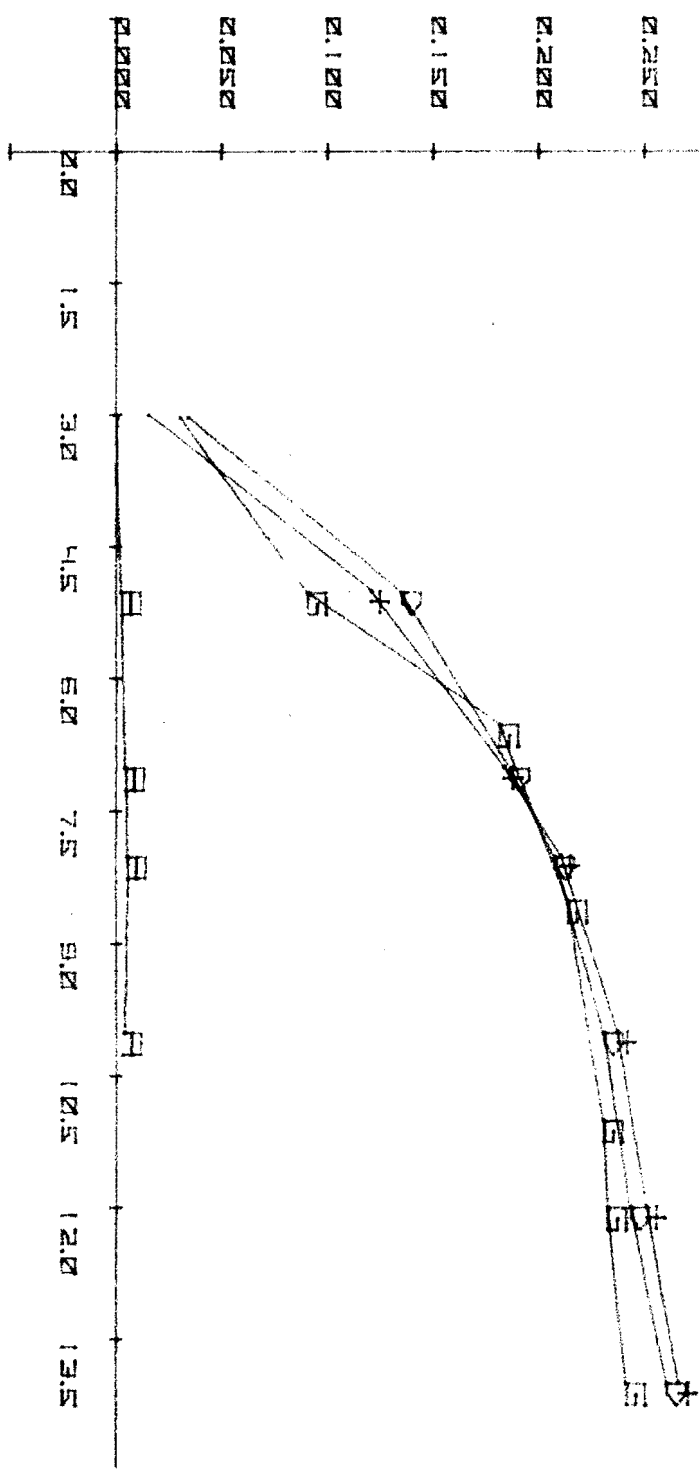
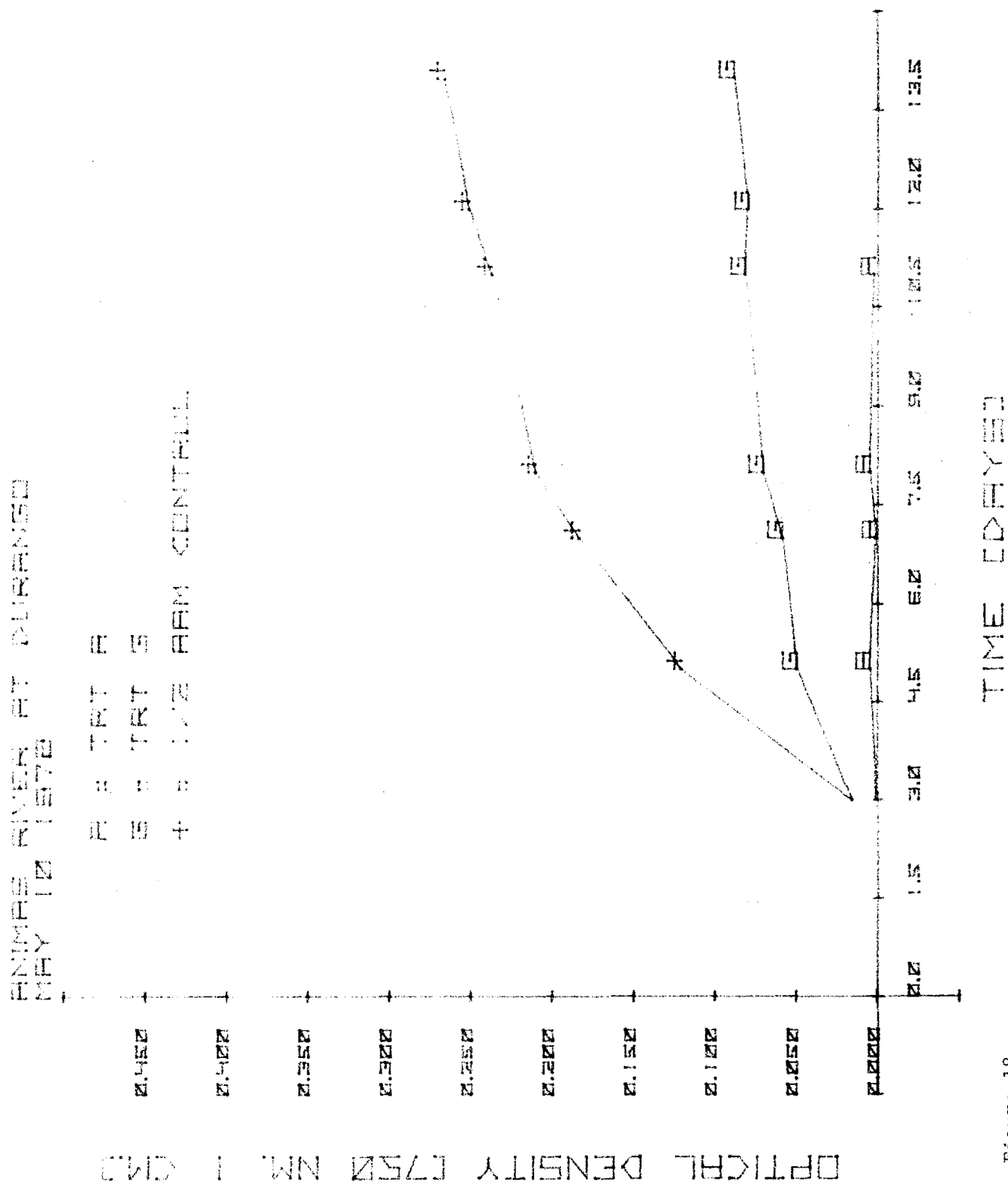


Figure 17.

TIME (MINUTES)

Figure 18.



MINNESOTA RIVER AT DURNING MAY 10, 1978

A = TRT A, B, C, E, F + EDTA
 D = TRT D + EDTA
 E = TRT E + EDTA
 + = 1/2 REM CONTROL + EDTA

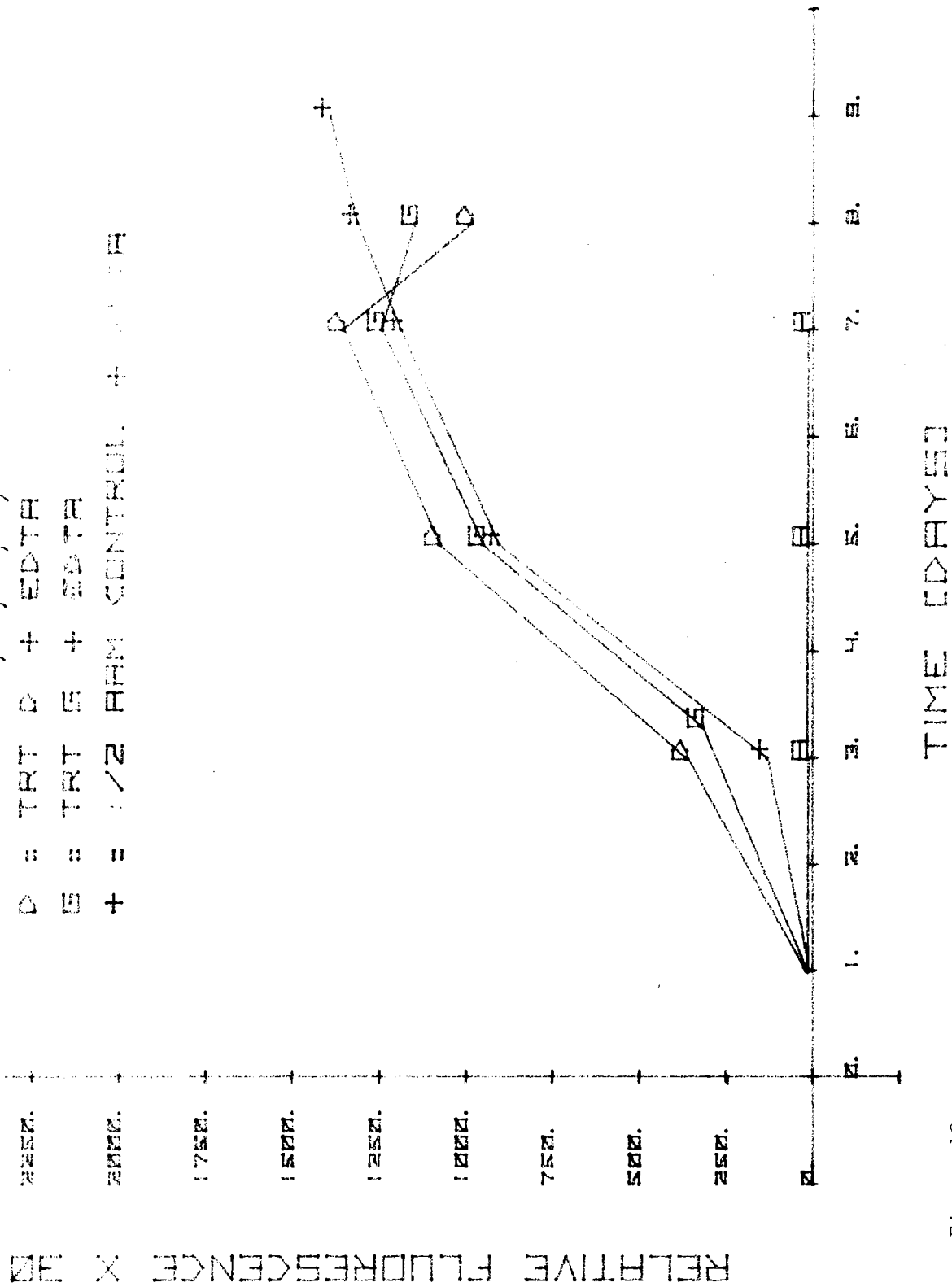


Figure 19.

2. Animas River at 32nd Street Bridge - Figure 21-40

Results of the algal assays at the 32nd Street Bridge site suggested a situation similar to that found at the Durango site. The sample was limited by both nitrogen and phosphorus at each sampling date, with phosphorus being the most limiting. A heavy metal toxicity problem was also indicated. Due to a definite increase in algal growth upon treatment with EDTA as compared to no EDTA addition, heavy metals were definitely pinpointed as the cause of toxicity. However, no heavy metals data were available for this site upon which to base any assumptions concerning particular metals involved. The close proximity of the Animas River sites (only several miles apart) leads one to the conclusion that zinc played a role in toxicity at the 32nd Street Bridge site as well as at the Animas site. The degree of toxicity at this site didn't follow the same pattern observed at the Durango site, however. The toxicity level varied from month to month with no definite pattern. This fact coupled with no available heavy metals data makes it impossible to draw any valid assumptions concerning the cause of toxicity on the Animas River at 32nd Street Bridge.

The low indigenous concentration of nitrogen and phosphorus as determined by chemical analysis was verified by algal bioassay. This extremely low nutrient level along with the heavy metal toxicity point in the direction of an oligotrophic to mesotrophic condition at this site on the Animas River.

ANIMHS RIVER AT 12ND STREET BRIDGE

SEPTEMBER 8 1977

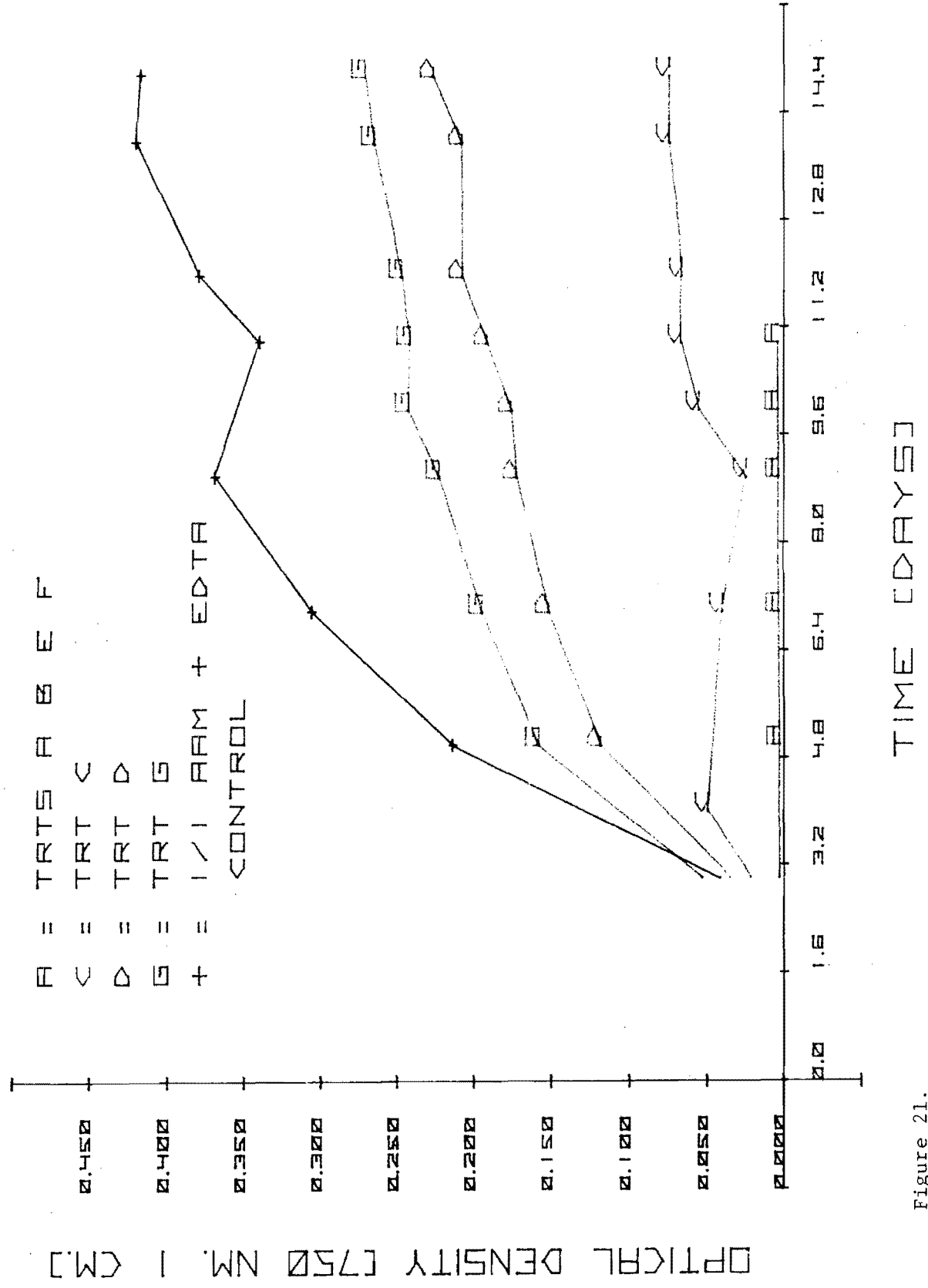


Figure 21.

ANIMAS RIVER AT 32ND STREET BRIDGE
SEPTEMBER 8 1977

OPTICAL DENSITY [750 NM. 1 CM.]

1 =
+ = 1/1 AM CONTROL

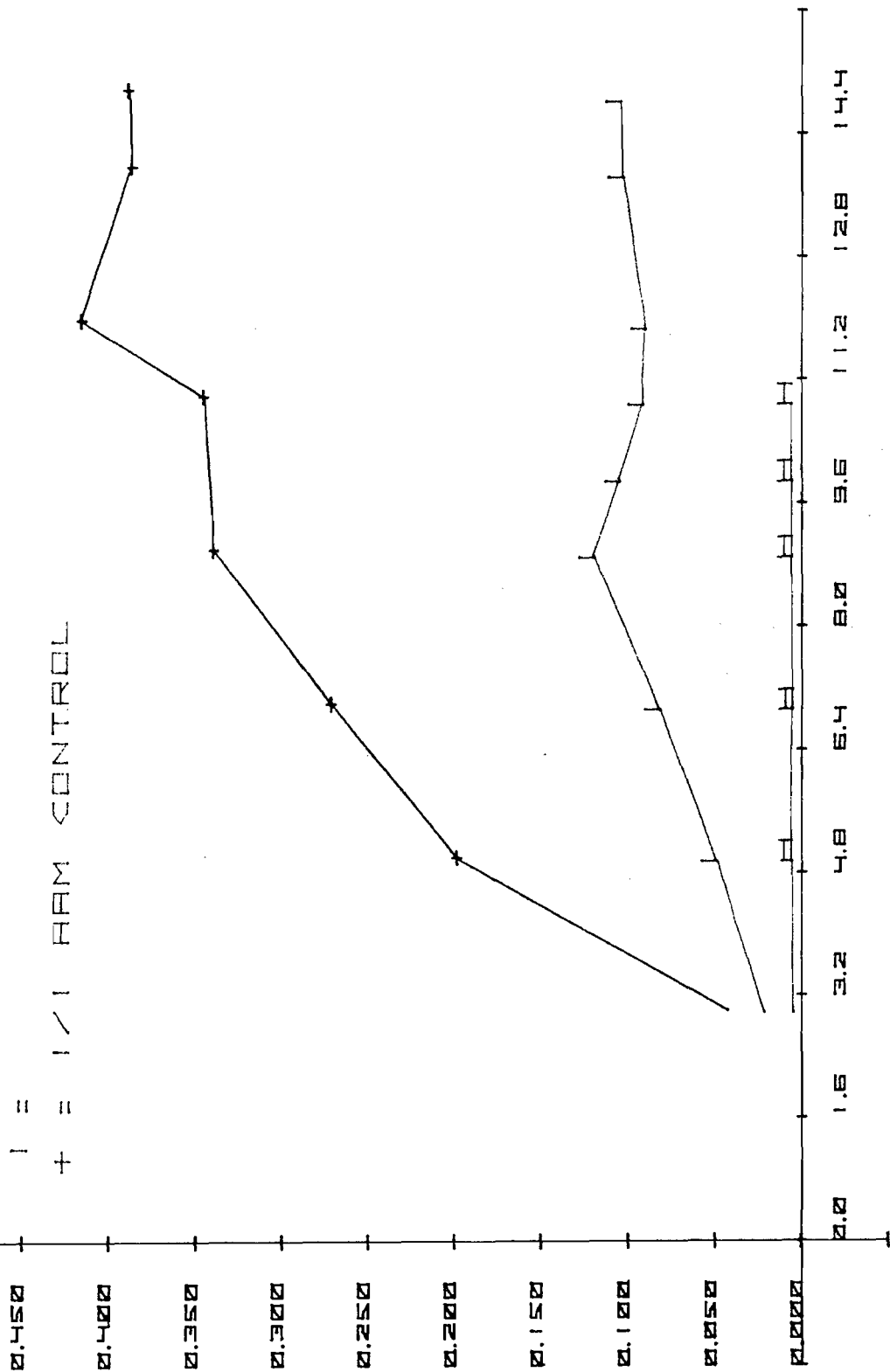


Figure 22.

TIME [DAYS]

ANIMAS RIVER AT 32ND STREET BRIDGE

SEPTEMBER, 8 1977

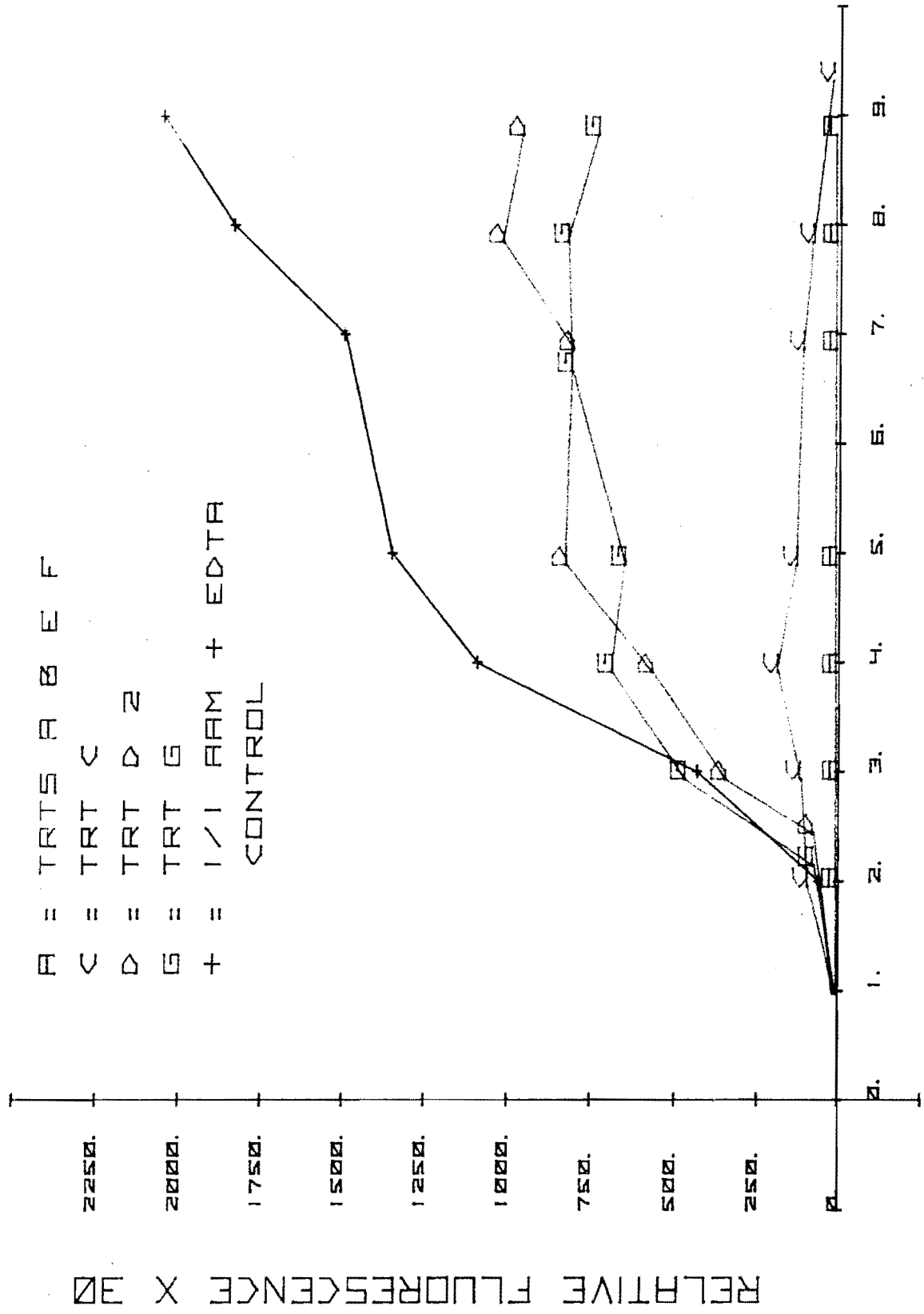


Figure 23.

ANIMAS RIVER AT 32ND STREET BRIDGE

SEPTEMBER 8 1977

H = TRT H

I = TRT I

+ = 1/1 RAM CONTROL

RELATIVE FLUORESCENCE X 30

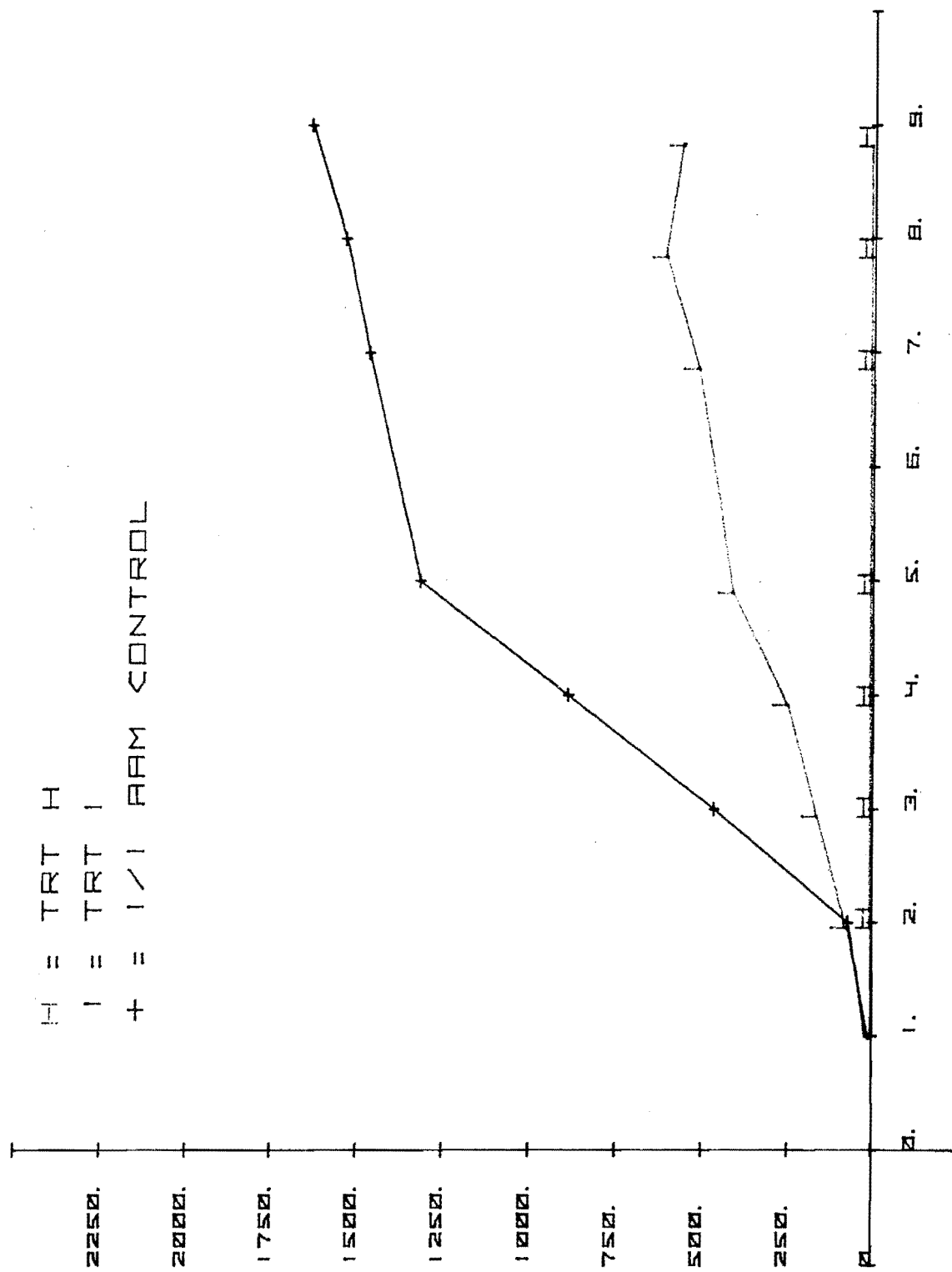


Figure 24.

TIME [DAYS]

ANIMAS RIVER AT 32ND STREET BRIDGE
NOVEMBER 29 1977.

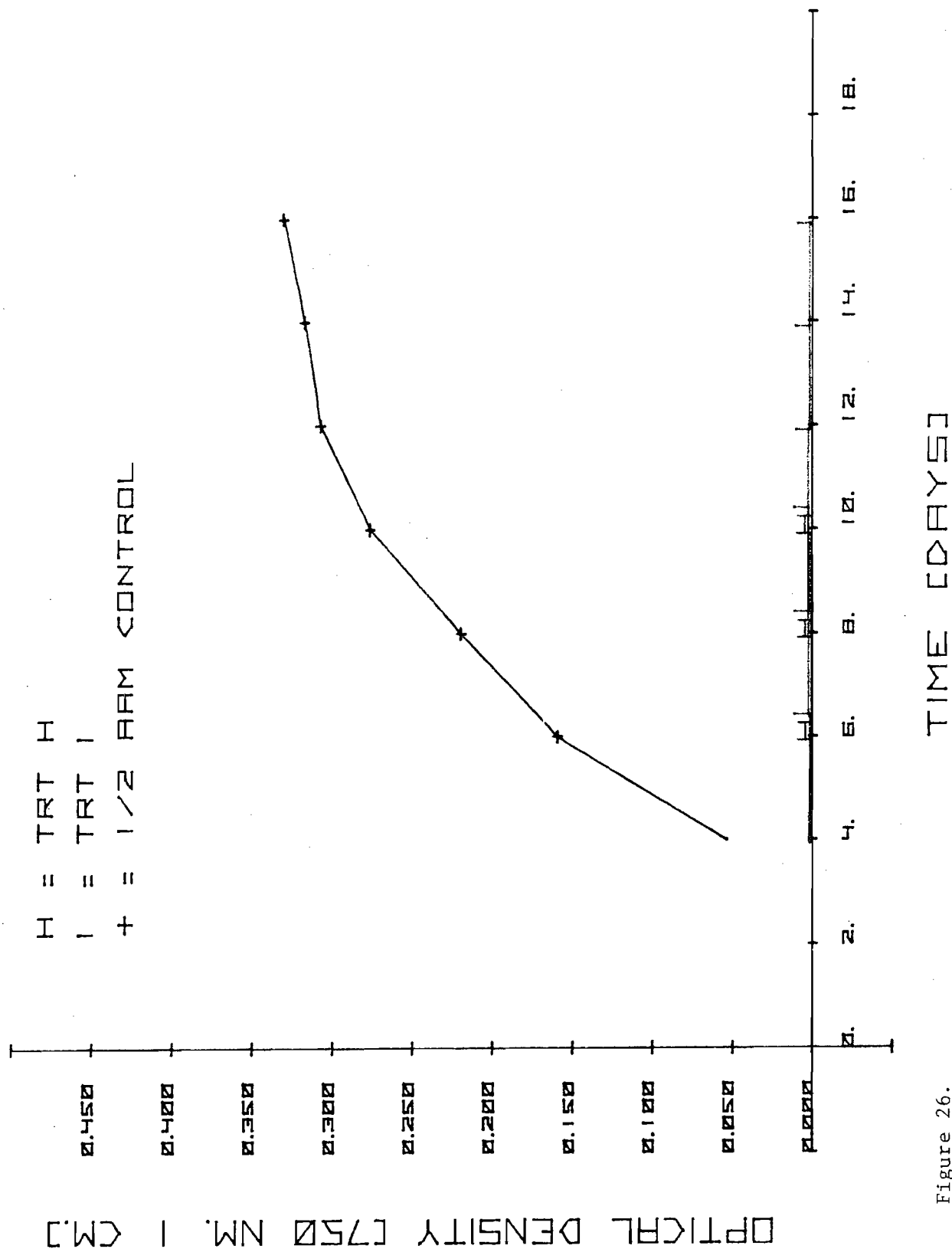


Figure 26.

ANIMAS RIVER AT 32ND STREET BRIDGE
NOVEMBER 29 1977

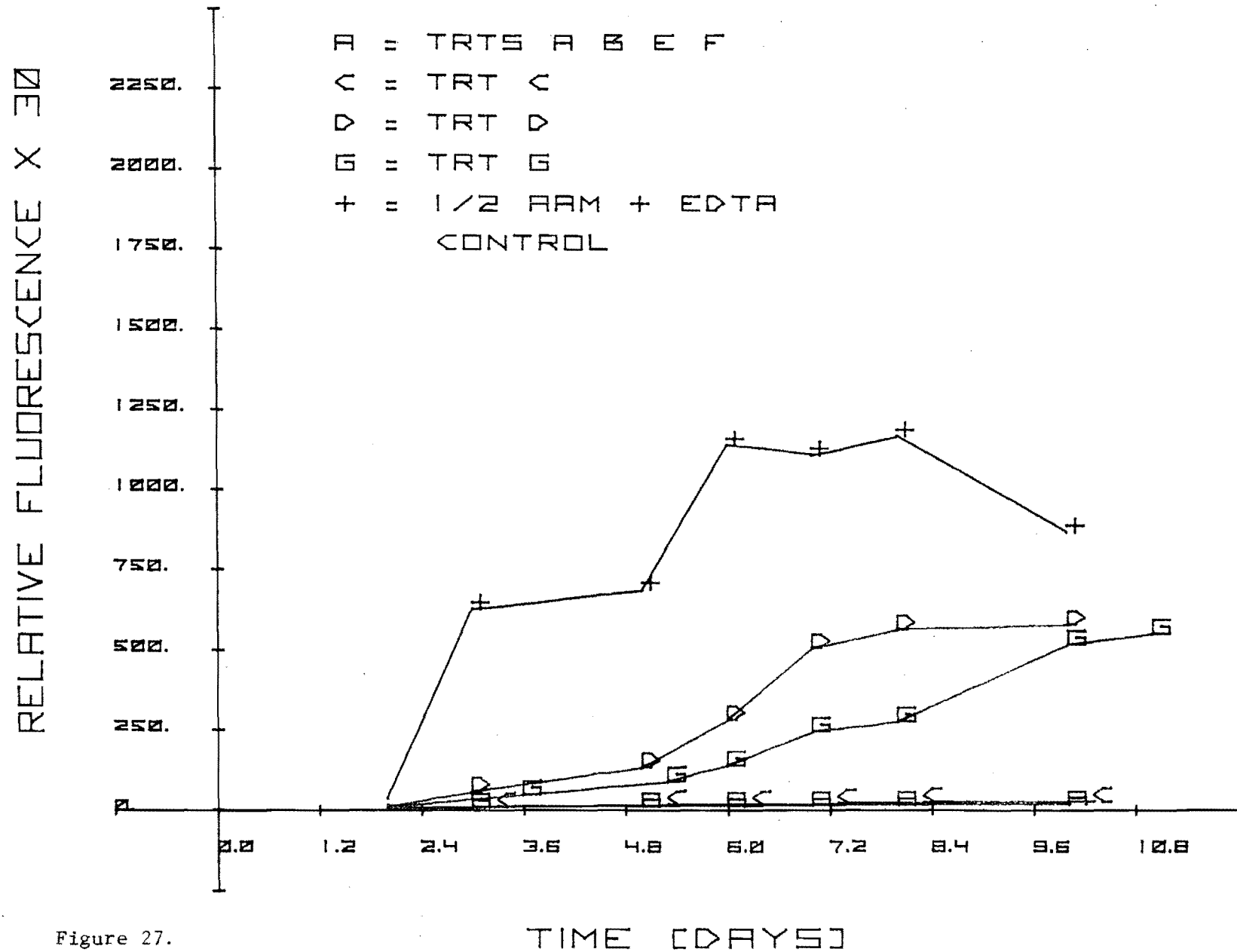


Figure 27.

ANIMAS RIVER AT 32ND STREET BRIDGE
NOVEMBER 29, 1977.

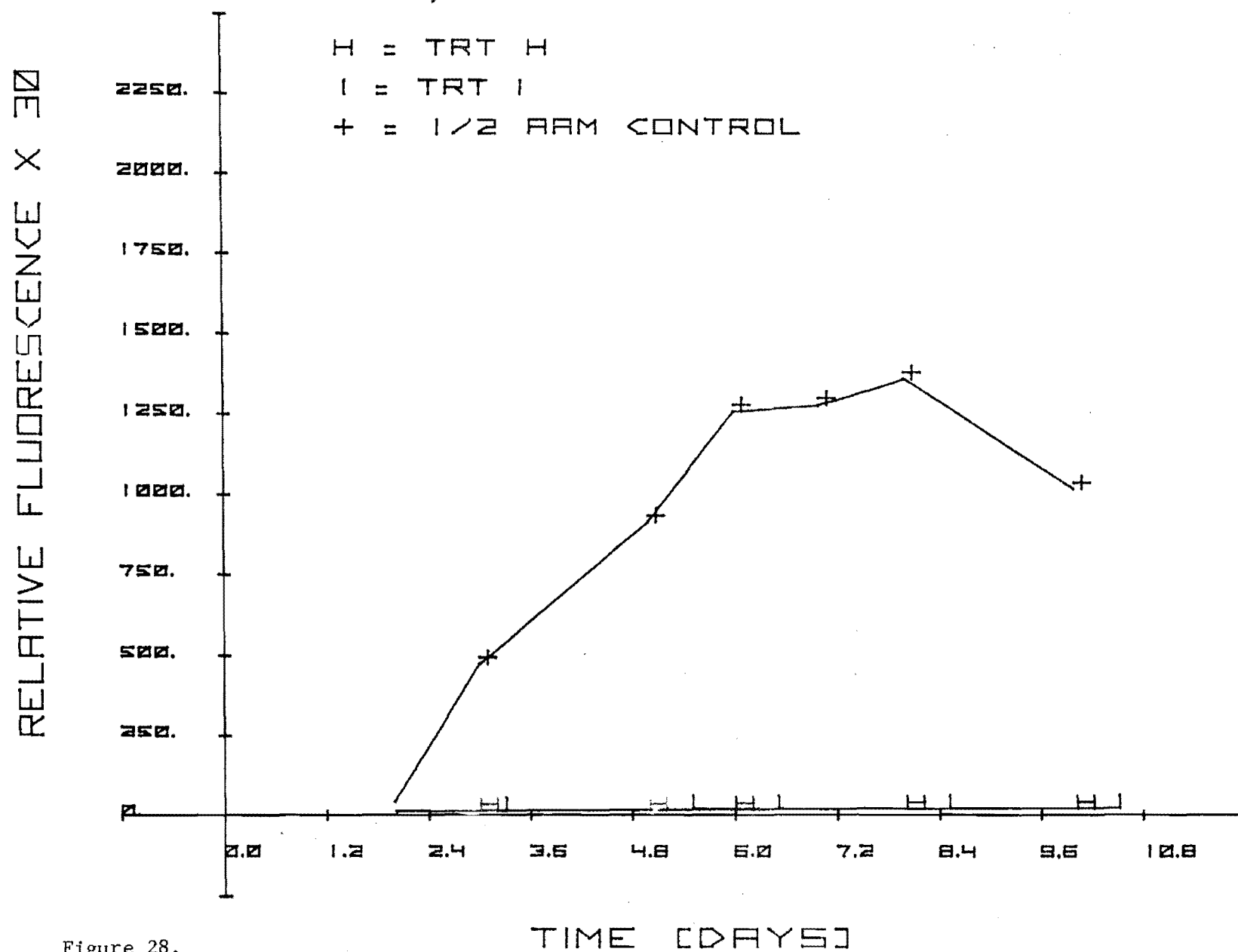
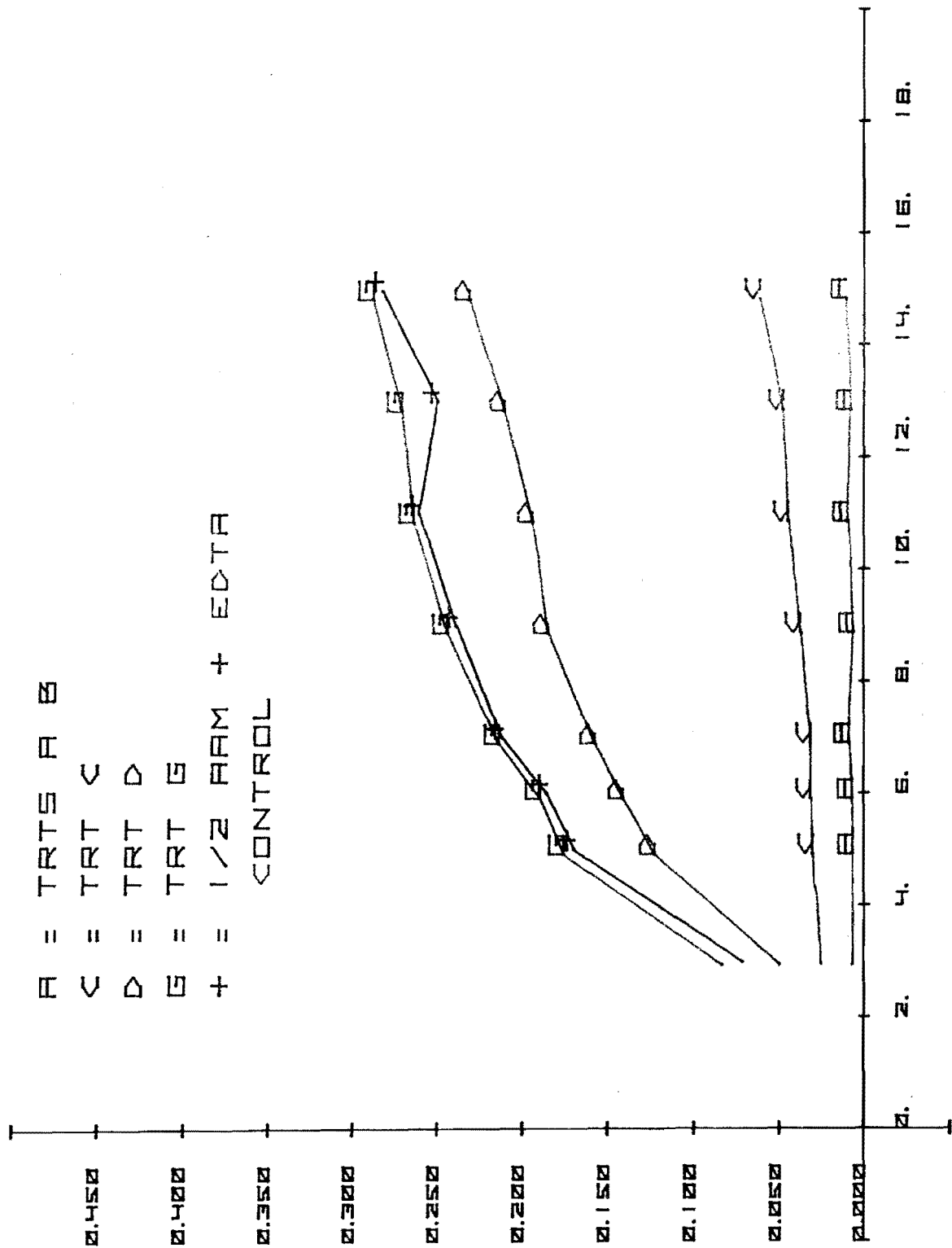


Figure 28.

ANIMAS RIVER AT 32ND STREET BRIDGE
JANUARY 9 1978

A = TRTS A B
C = TRT C
D = TRT D
E = TRT E
+ = 1/2 ARM + EDTA
CONTROL

OPTICAL DENSITY [750 NM. 1 CM.]



TIME [DAYS]

Figure 29.

ANIMAS RIVER AT 32ND STREET BRIDGE.
JANUARY 9 1978

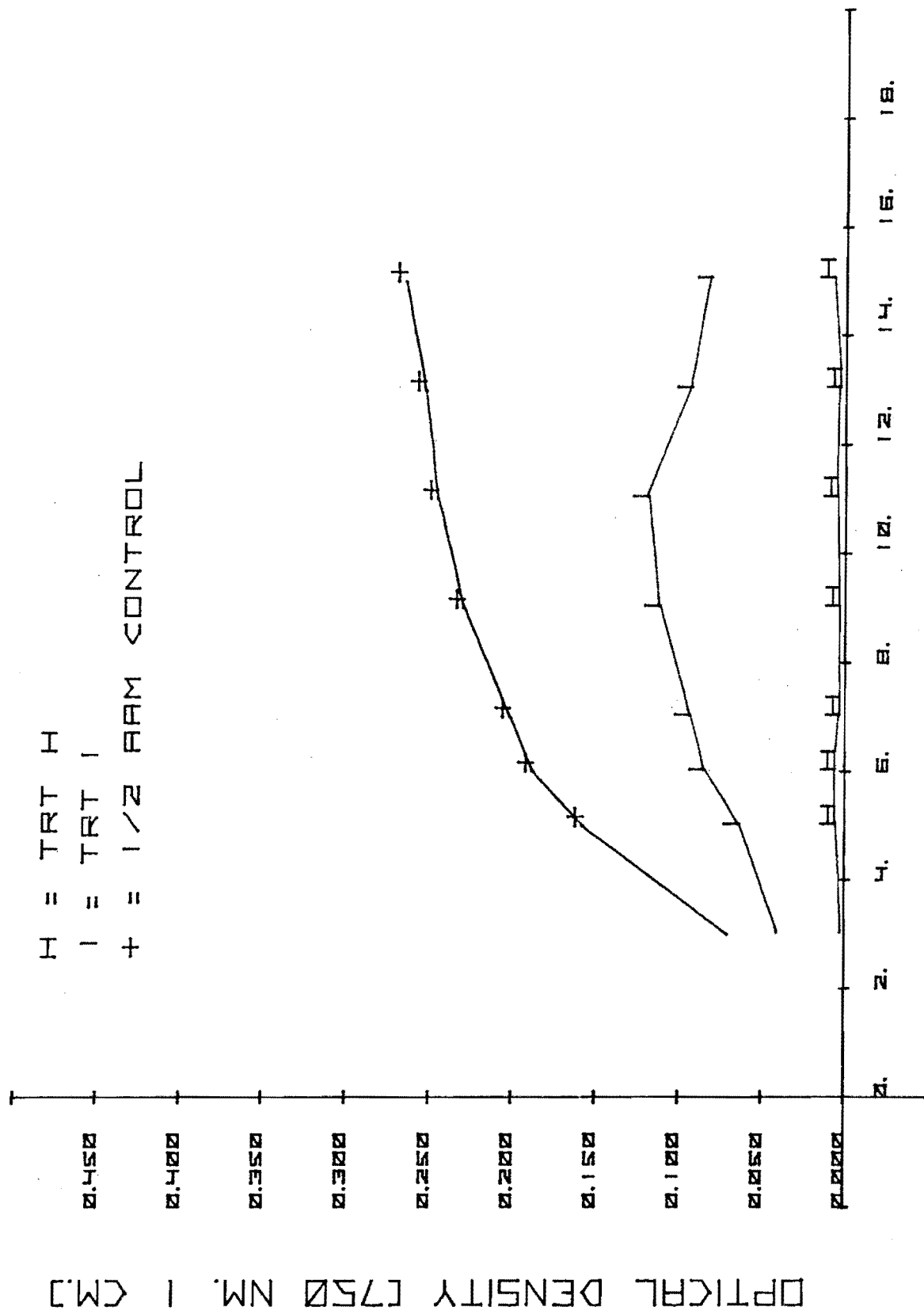


Figure 30.

ANIMAS RIVER AT 32ND STREET BRIDGE
JANUARY 9 1978

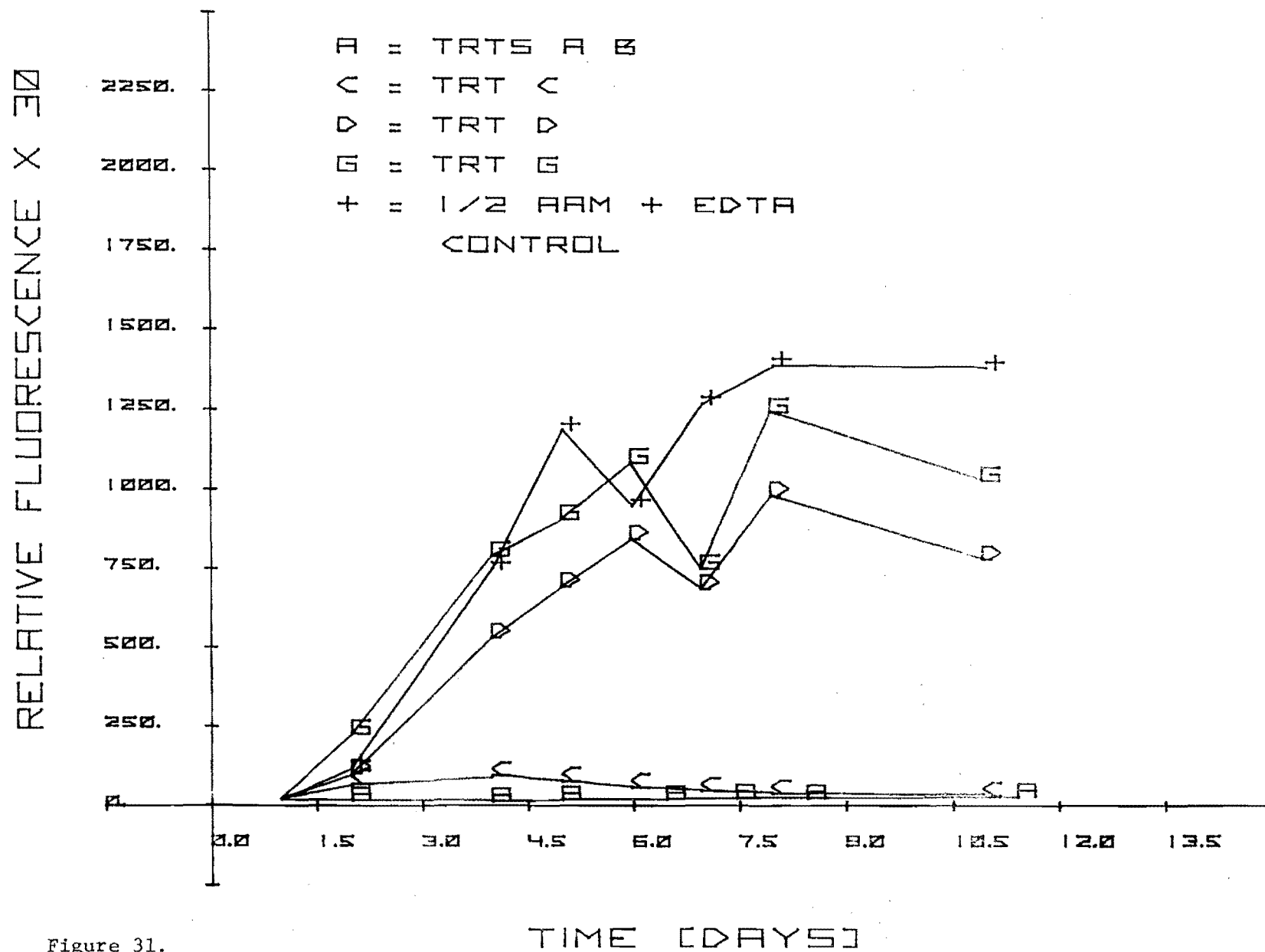


Figure 31.

ANIMAS RIVER AT 32ND STREET BRIDGE
JANUARY 9 1978

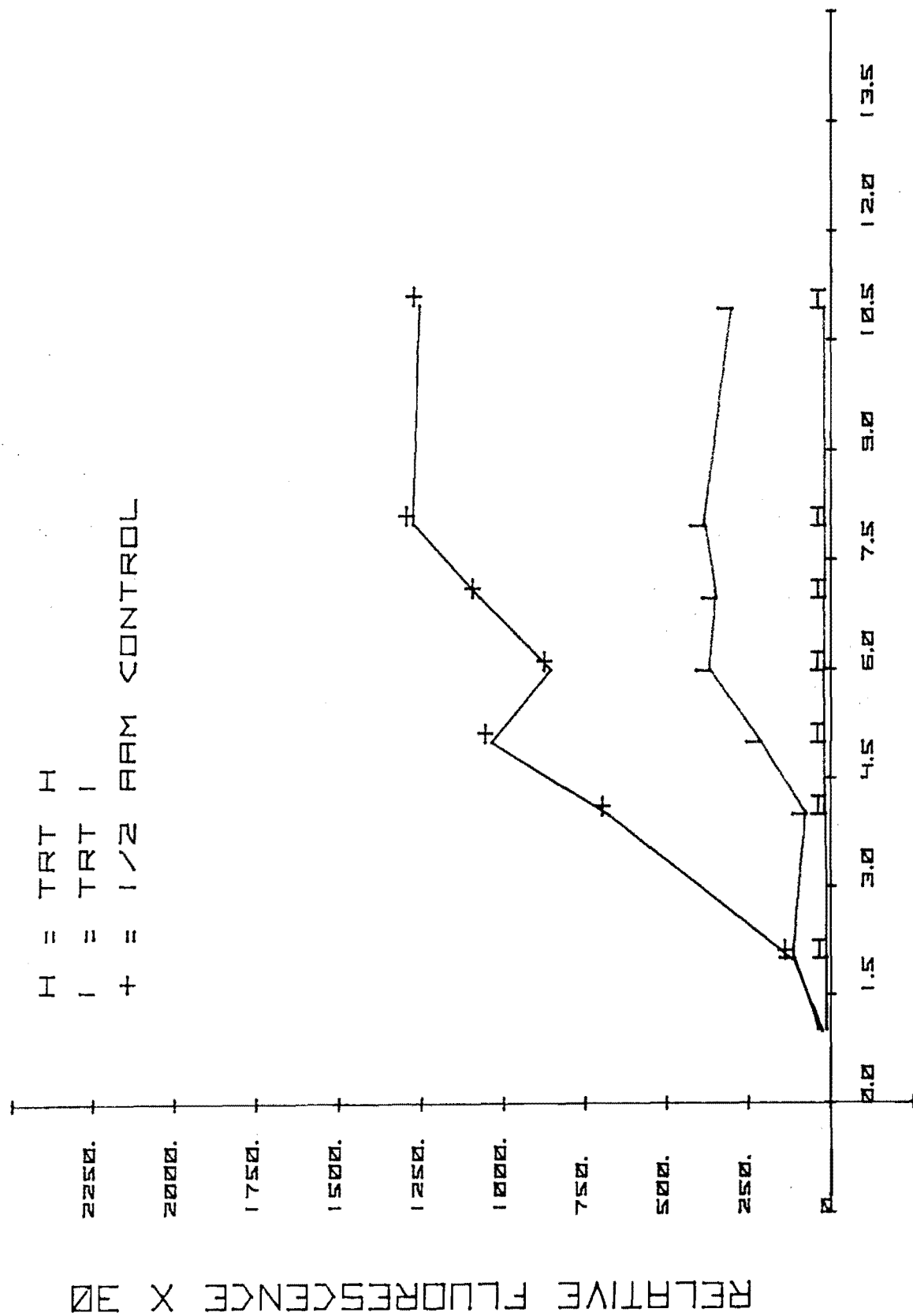


Figure 32.

TIME COPIES

ANIMAS RIVER AT 32ND STREET BRIDGE

MARCH 8 1978

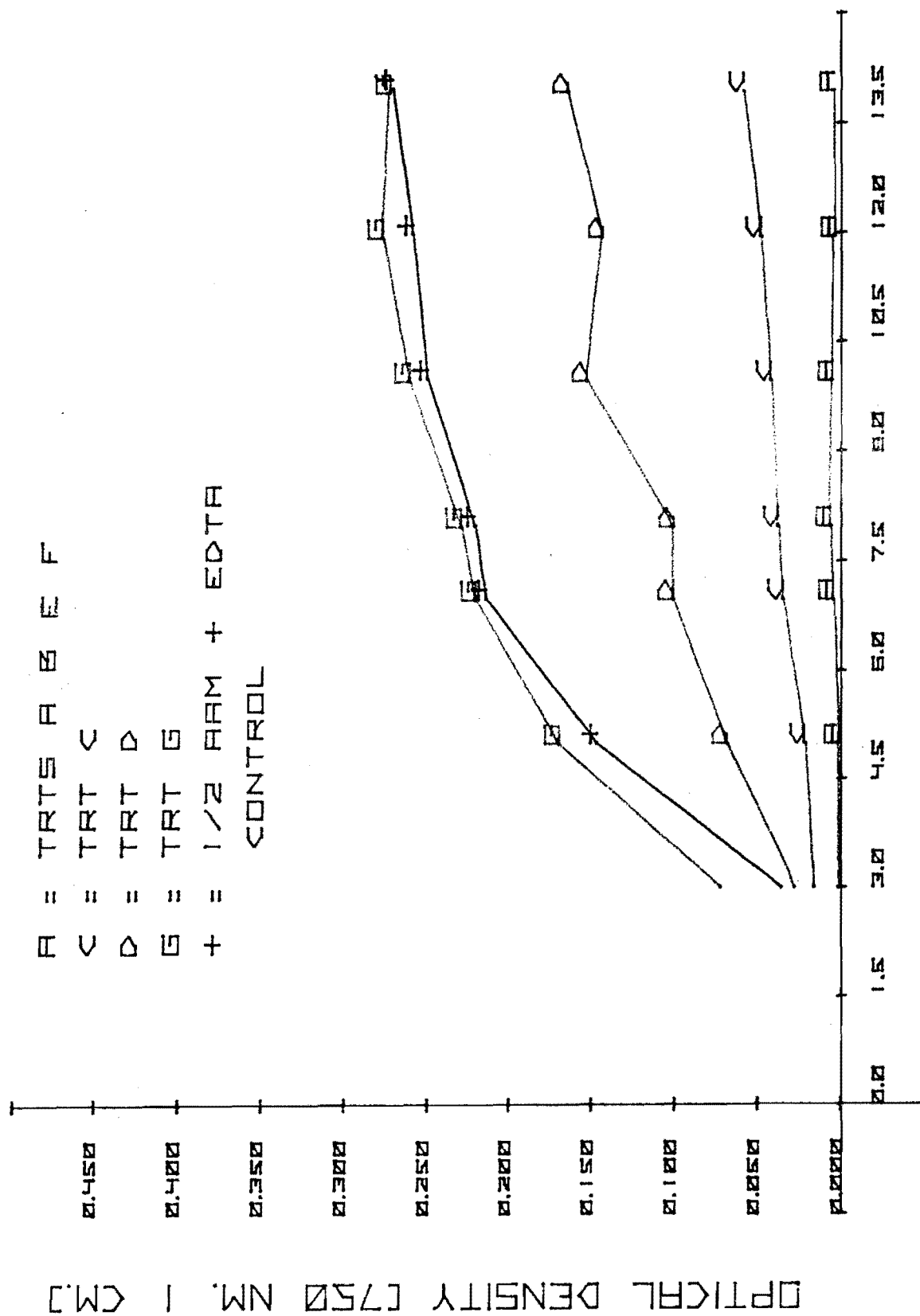


Figure 33.

ANIMAS RIVER AT 32ND STREET BRIDGE.

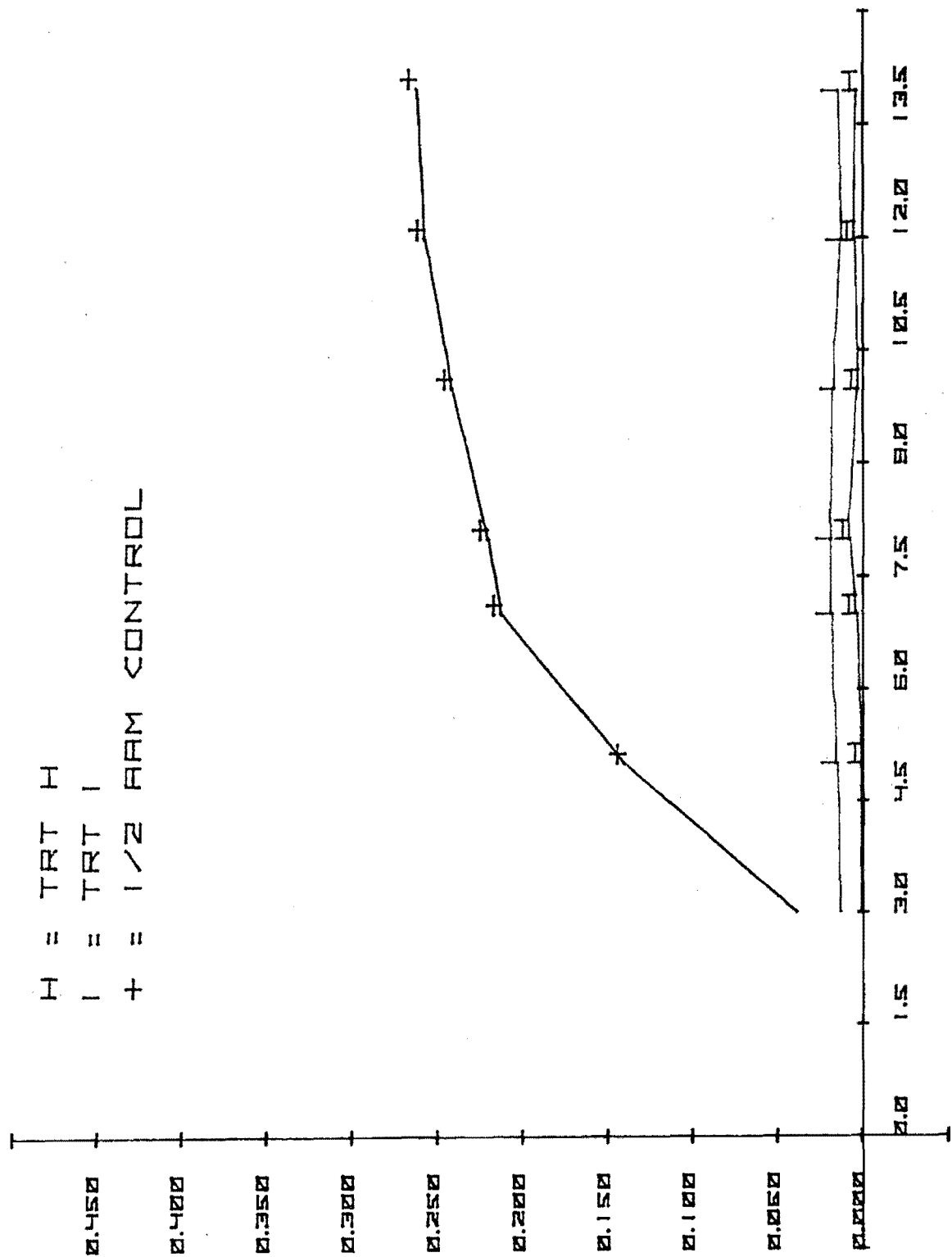
MARCH 8 1978

H = TRT H

I = TRT I

+ = 1/2 AM CONTROL

OPTICAL DENSITY [750 NM. 1 CM.]



TIME (DAYS)

Figure 34.

ANIMAS RIVER AT 32ND STREET BRIDGE
MARCH 8 1978

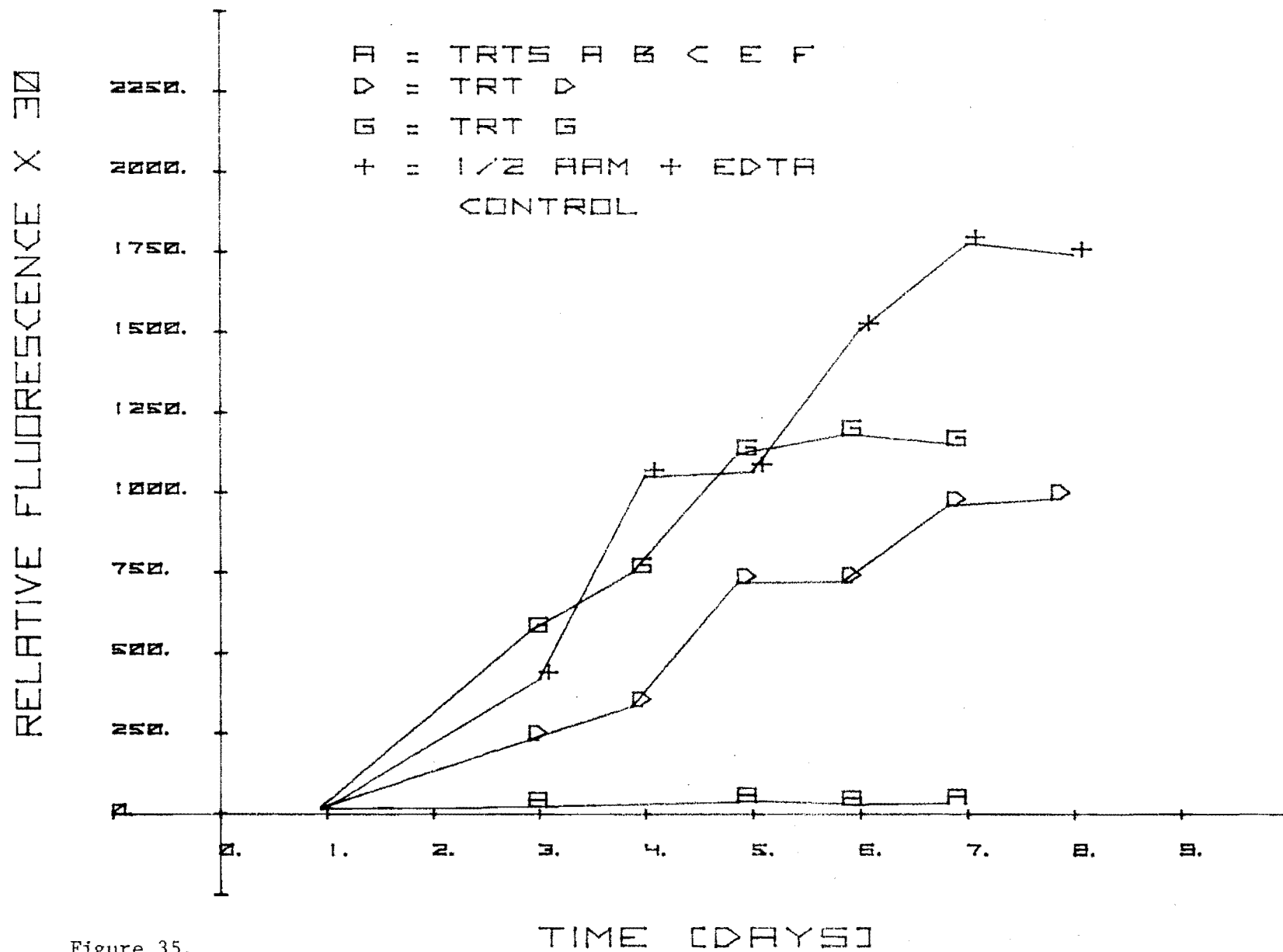


Figure 35.

ANIMAS RIVER AT 32ND STREET BRIDGE
MARCH 8 1978

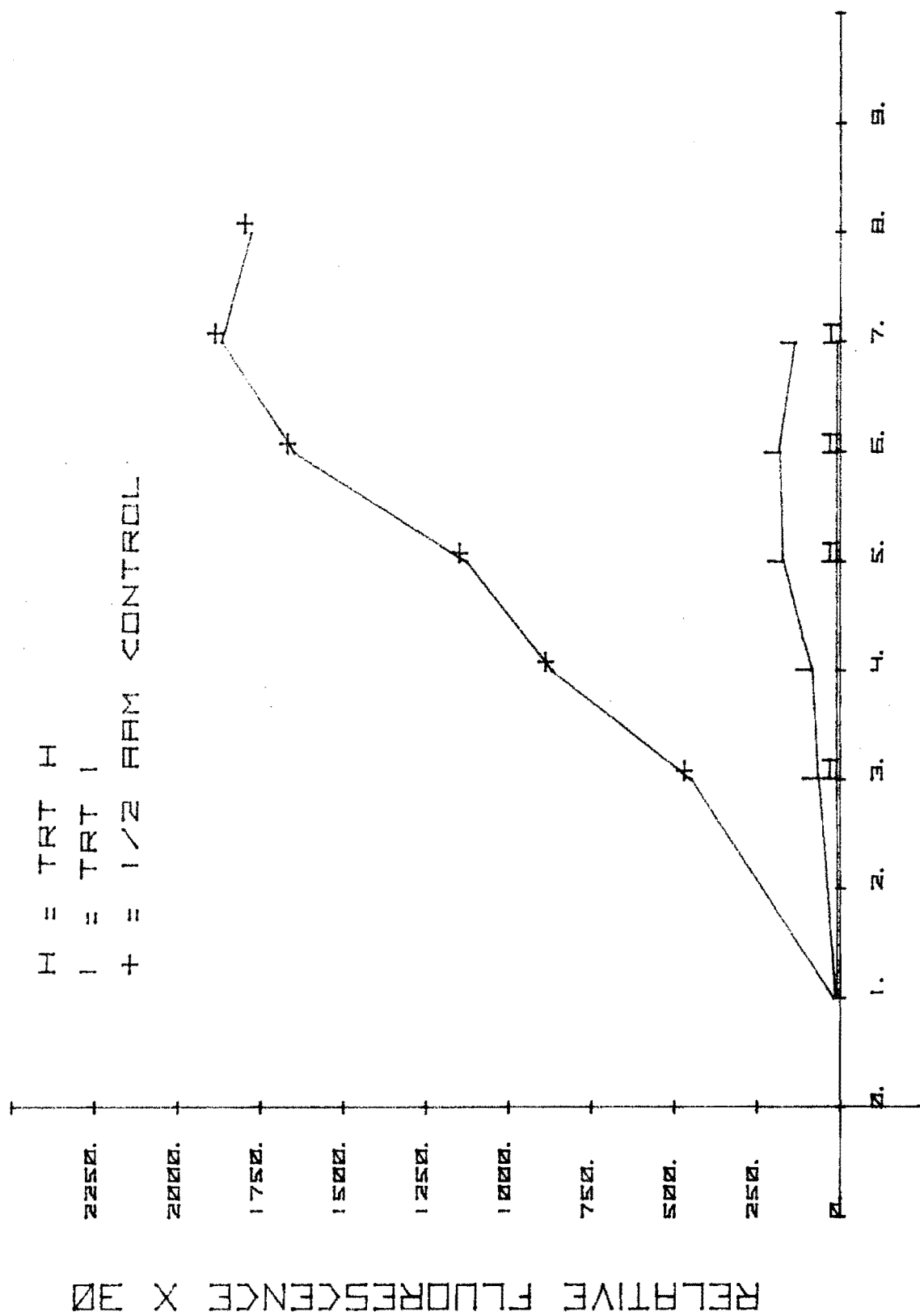


Figure 36.

ANIMAS RIVER AT 32ND ST. BRIDGE
MAY 10, 1978

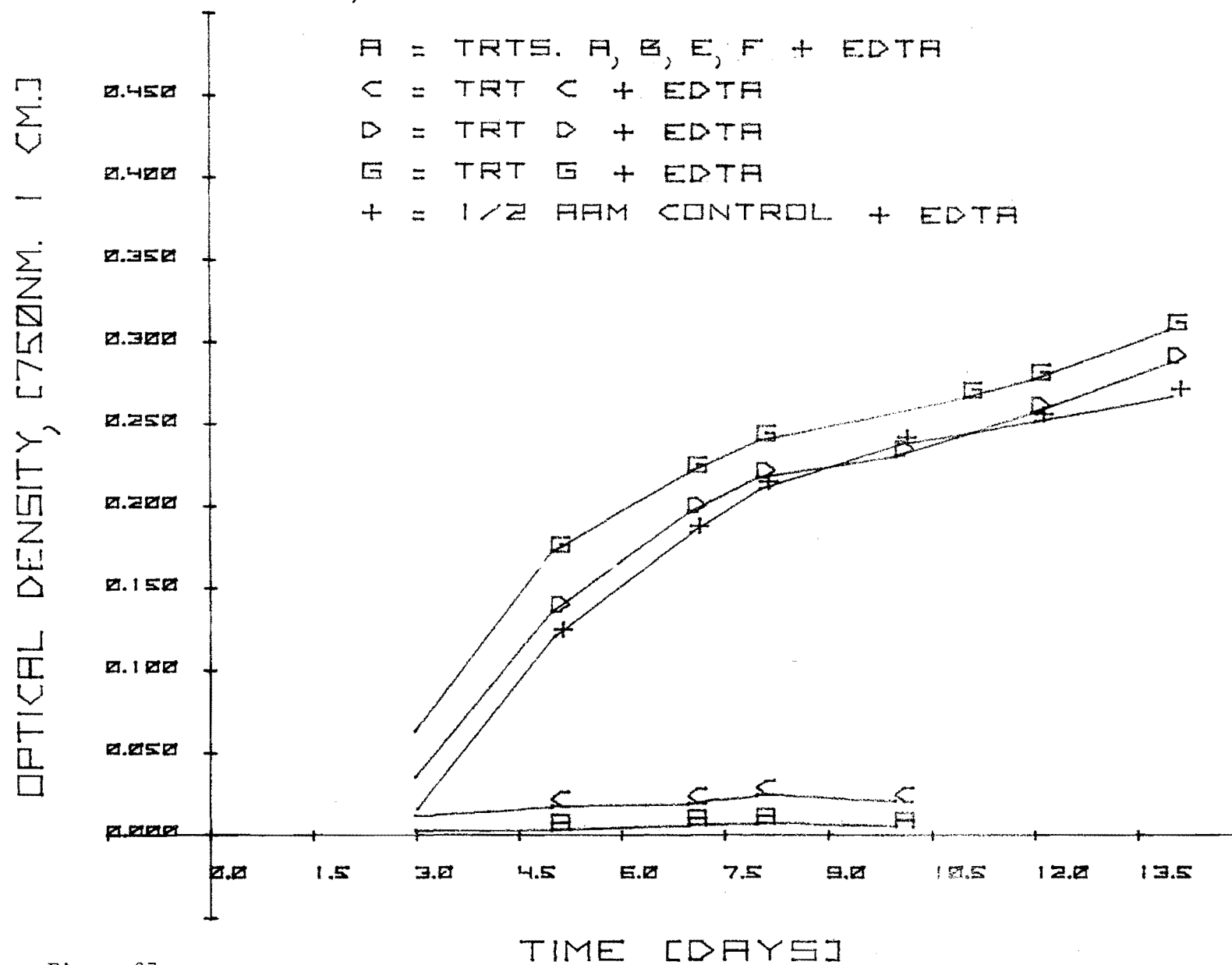


Figure 37.

ANIMAS RIVER AT 32ND ST. BRIDGE
MAY 10 1978

A = TRT A
G = TRT G
+ = 1/2 HAM CONTROL

OPTICAL DENSITY [750 NM. 1 CM.]

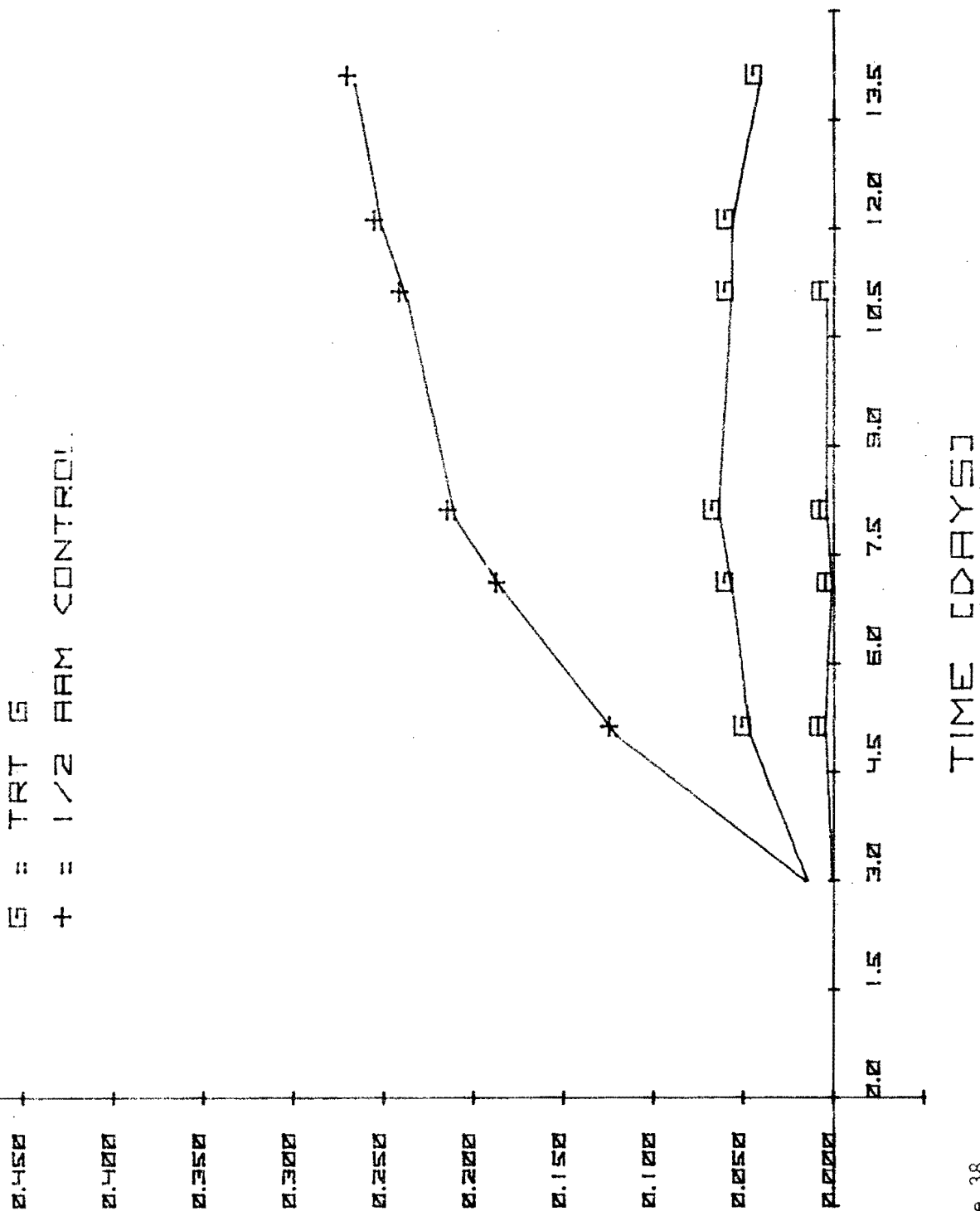


Figure 38.

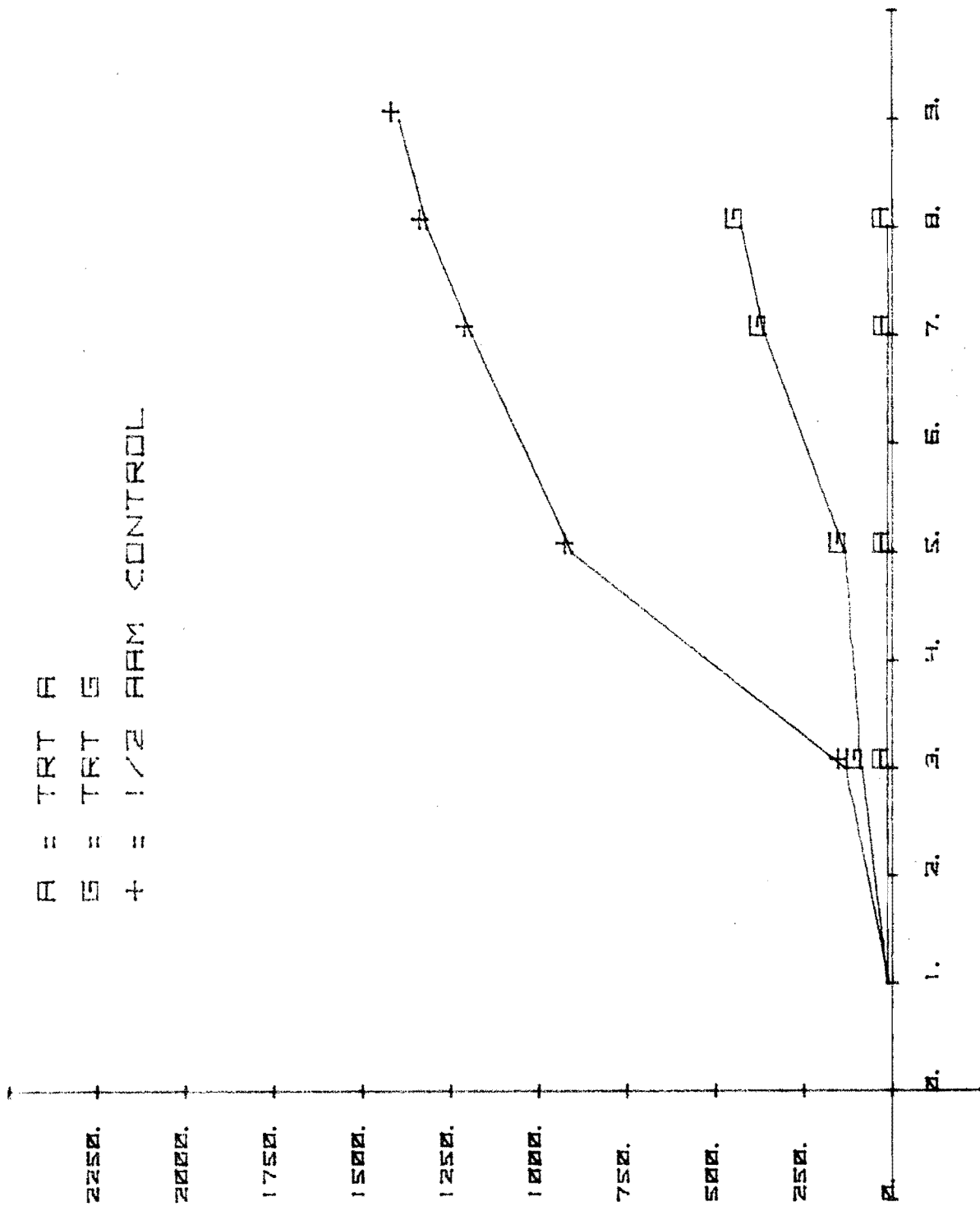
ANIMAS RIVER AT 32ND ST. BRIDGE
MAY 10 1978 2

A = TRT A

G = TRT G

+ = 1/2 ARM CONTROL

RELATIVE FLUORESCENCE X 30



TIME [DAYS]

Figure 40.

3. La Plata River at Colorado/New Mexico Border
Figures 41-54

Chemical analysis as well as bioassay indicated phosphorus limitation first and then nitrogen limitation during November, January and March. In May, 1978 the sample was simultaneously limited by nitrogen and phosphorus. The May bioassay also showed a eutrophic response in the sample with no treatment as opposed to an oligotrophic response in all previous bioassays. The results of chemical analysis (Table III-1) indicated a greater concentration of phosphorus in May 1978 as compared to the previous months. The higher level of phosphorus created a near optimum M/P ratio of 22 and the result was greater productivity during the bioassay growth.

It should be noted that response upon addition of complete AAM during November, January, and March was significantly less than the AAM control. This was found to be the case regardless of whether EDTA was added or not. Often when this type of low algal response is observed it can be attributed to the hardness of the water and most specifically to the calcium concentration. Euster (1958) showed that photosynthetic activity caused removal of CO_2 resulting in higher pH. At the higher pH, the hardness precipitates, largely as CaCO_3 and/or coprecipitates $\text{Ca}_x(\text{PO}_4)_y$ compounds and heavy metals. Therefore, as the pH rises above pH 8.8, the CaCO_3 precipitate is formed removing CO_2 and other nutrients from the sample thus inhibiting algal growth and ultimately altering the results.

The calcium concentration in the La Plata River at State Line during November, January, March and May seemed to correlate well with the

observed algal response. In March, 1978 when algal response was the lowest, the calcium concentration (203 mg/l) was at the highest level observed during the entire monitoring period of 16 months. The calcium concentration in May, 1978 was at the lowest observed level (49 mg/l) and consequently the algal productivity was higher than during any other bioassay. The other algal bioassays at this site reflect the same trend. However, four bioassays do not provide a statistically sound basis from which to draw definite conclusions concerning calcium concentration as it relates to algal productivity in the La Plata River. Without further data, it may be speculated that the high calcium concentration created the lower than normal algal response.

LA PLATA RIVER AT COLO./N.M. BORDER
 NOVEMBER 29 1977

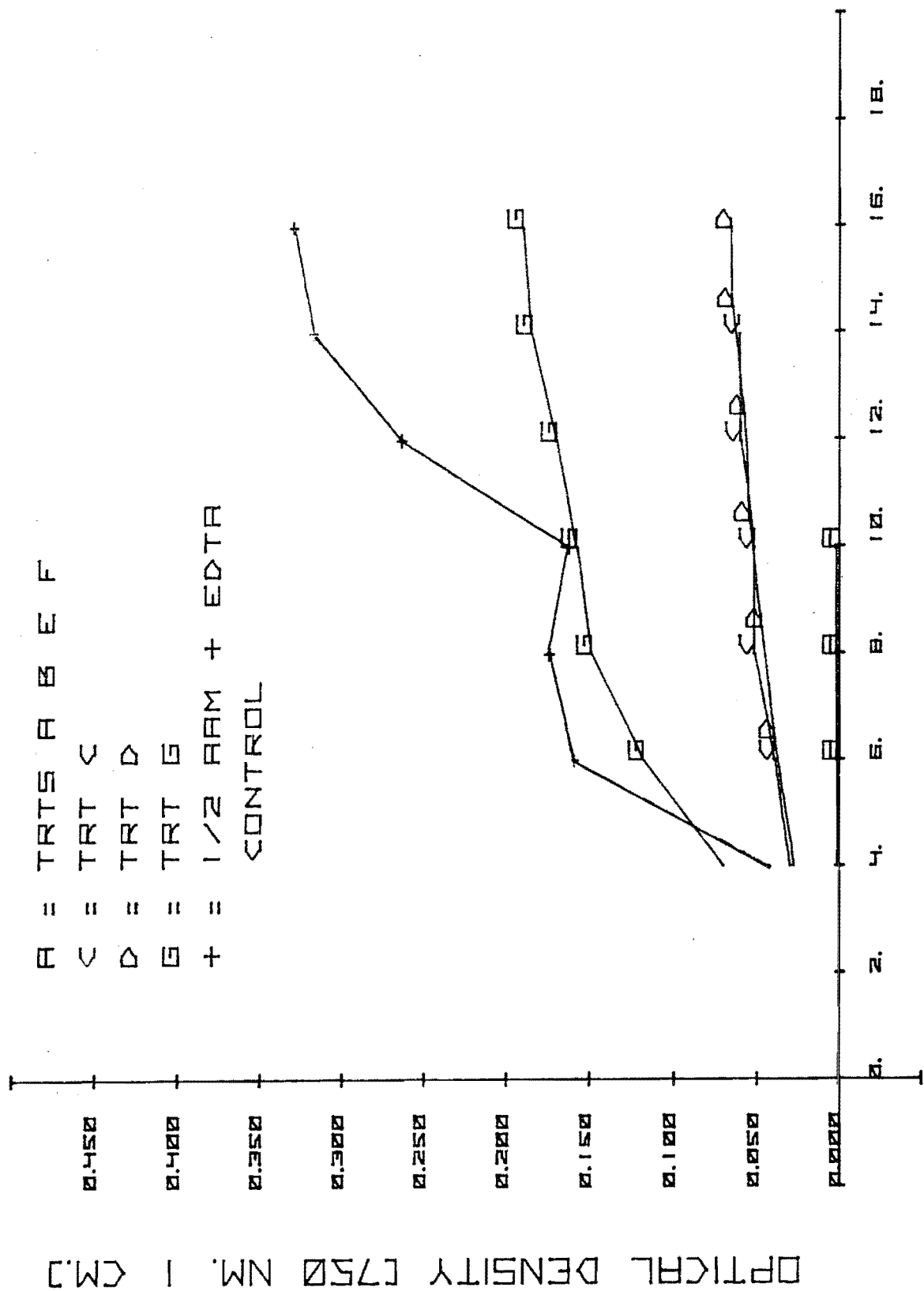


Figure 41.

LA PLATA RIVER AT COLD./N.M. BORDER
 NOVEMBER 29 1977.

H = TRT H
 I = TRT I
 + = 1/2 ARM CONTROL

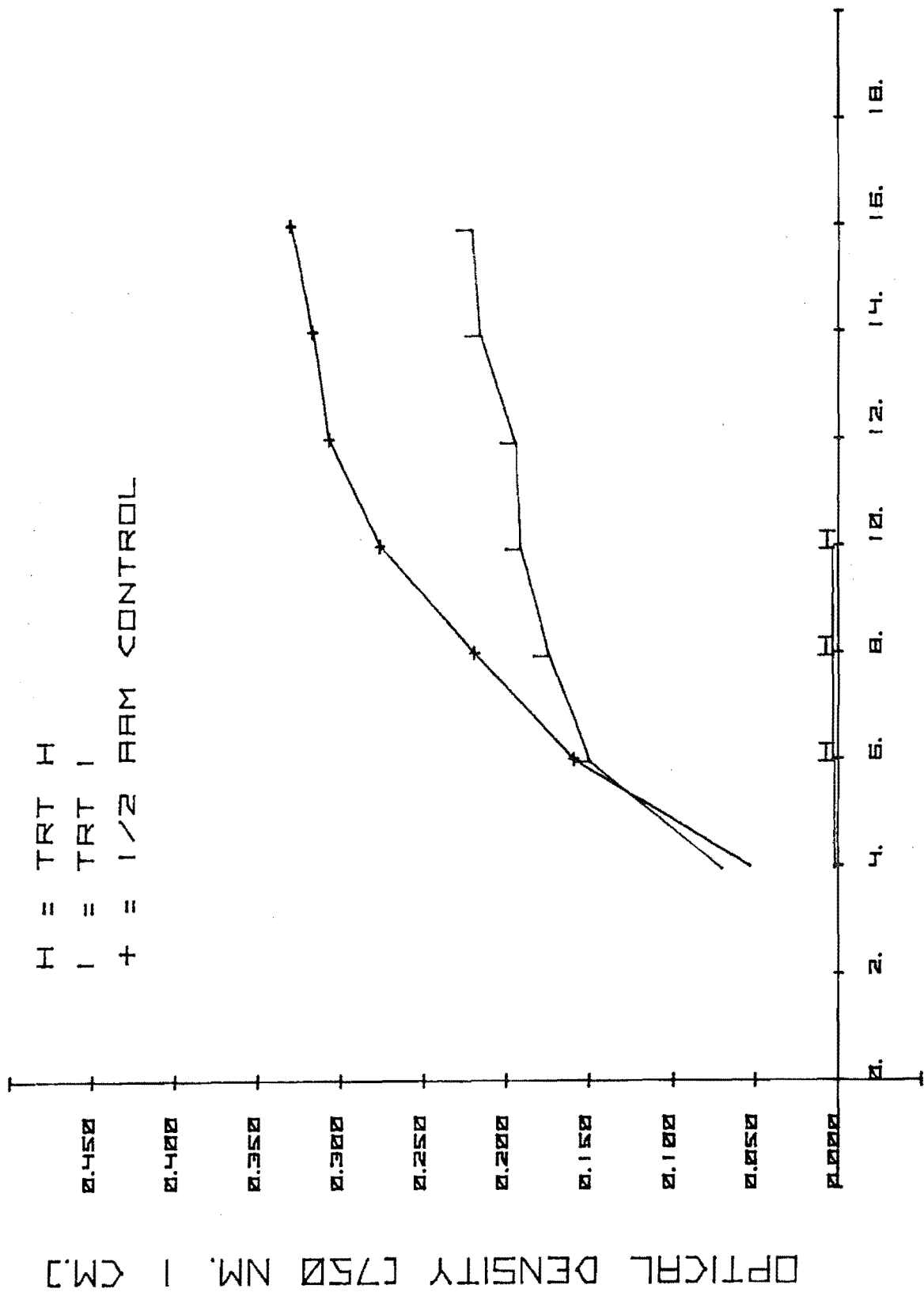


Figure 42.

LA PLATA RIVER AT COLD./N.M. BORDER
NOVEMBER 29, 1977.

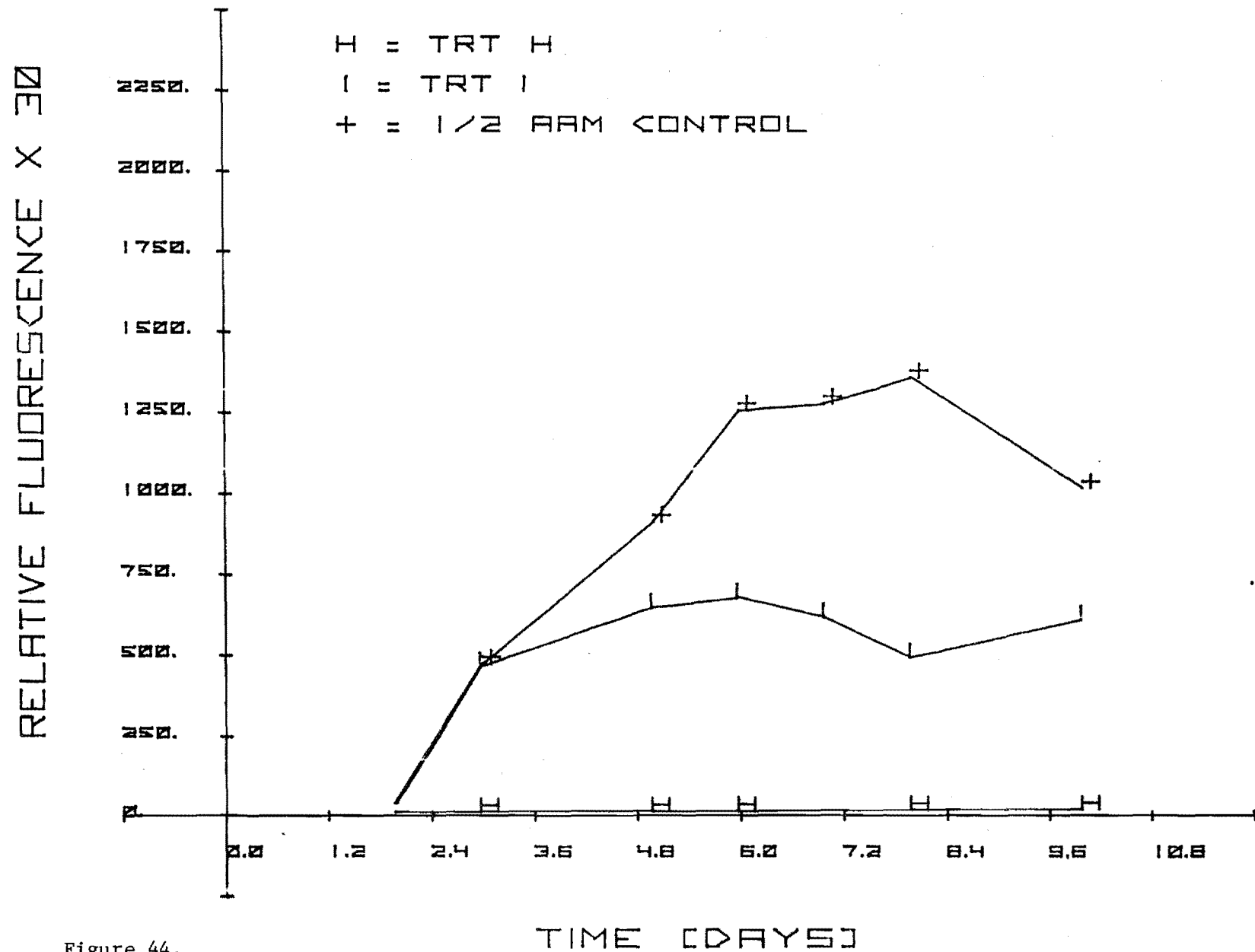


Figure 44.

LA PLATA RIVER AT COLO./N.M. BORDER
NOVEMBER 29 1977

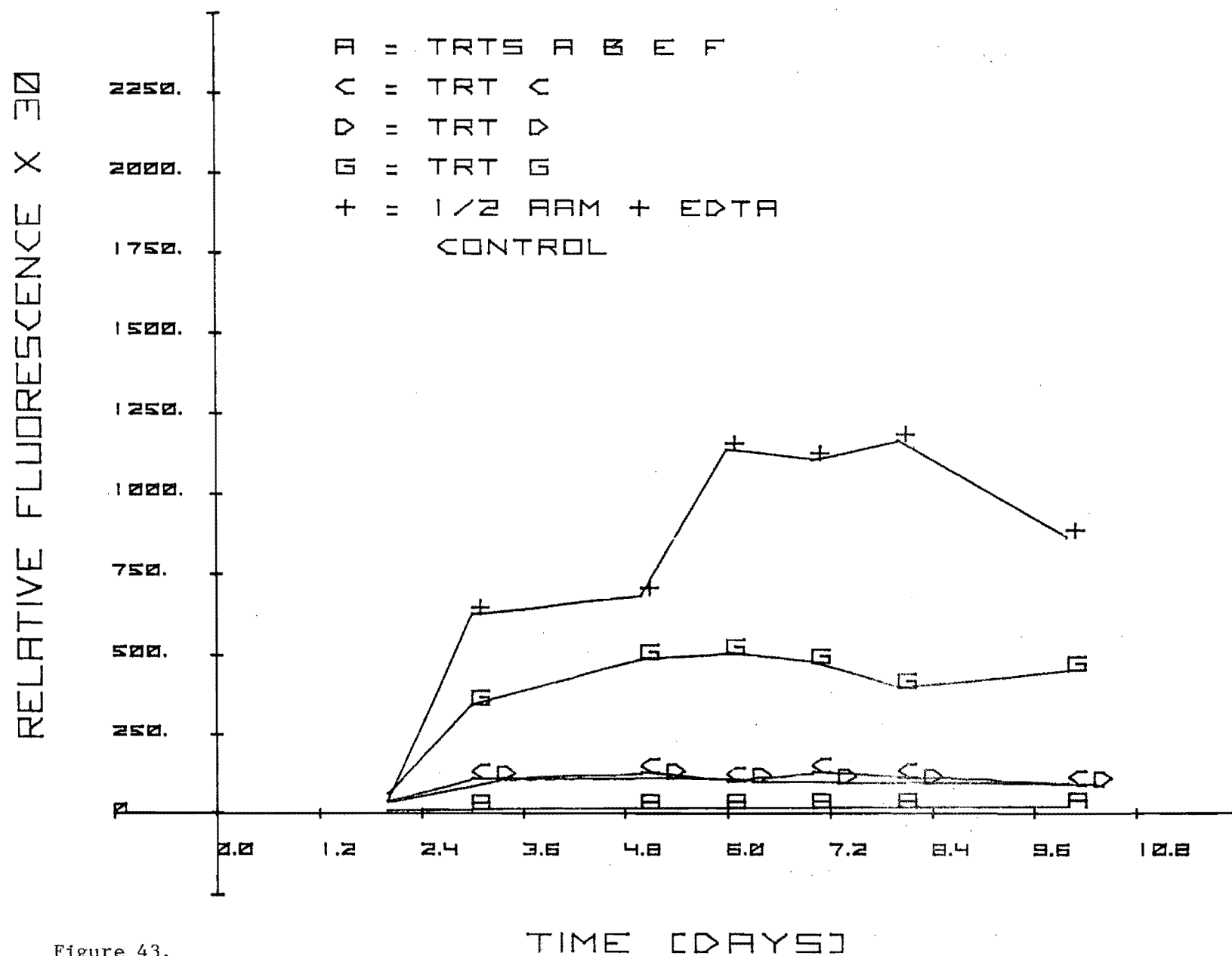
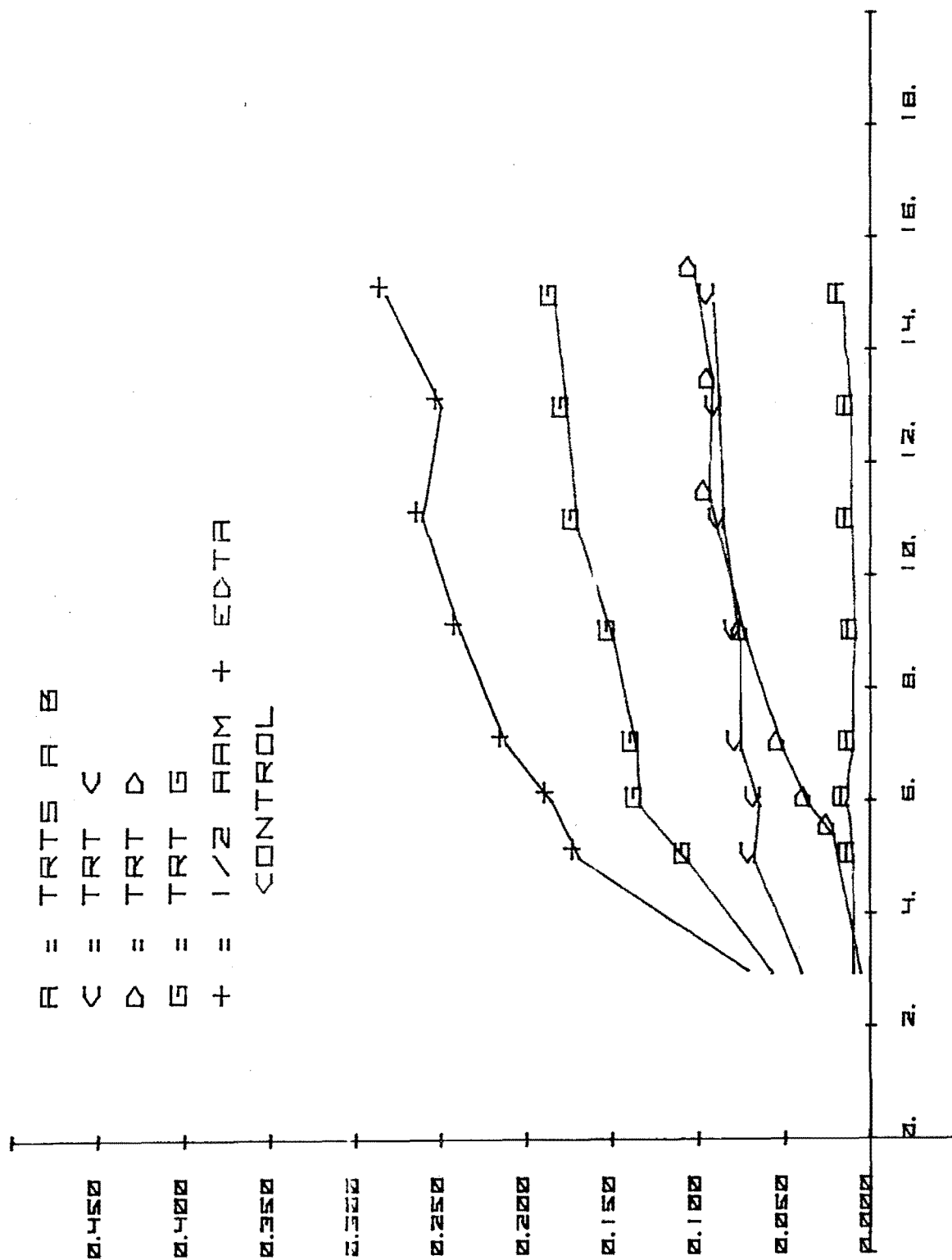


Figure 43.

LA PLATA RIVER AT COLO./N.M. BORDER
JANUARY 9 1978

A = TRTS A B
C = TRT C
D = TRT D
E = TRT E
+ = 1/2 ARM + EDTA
CONTROL

OPTICAL DENSITY [750 NM. 1 CM.]



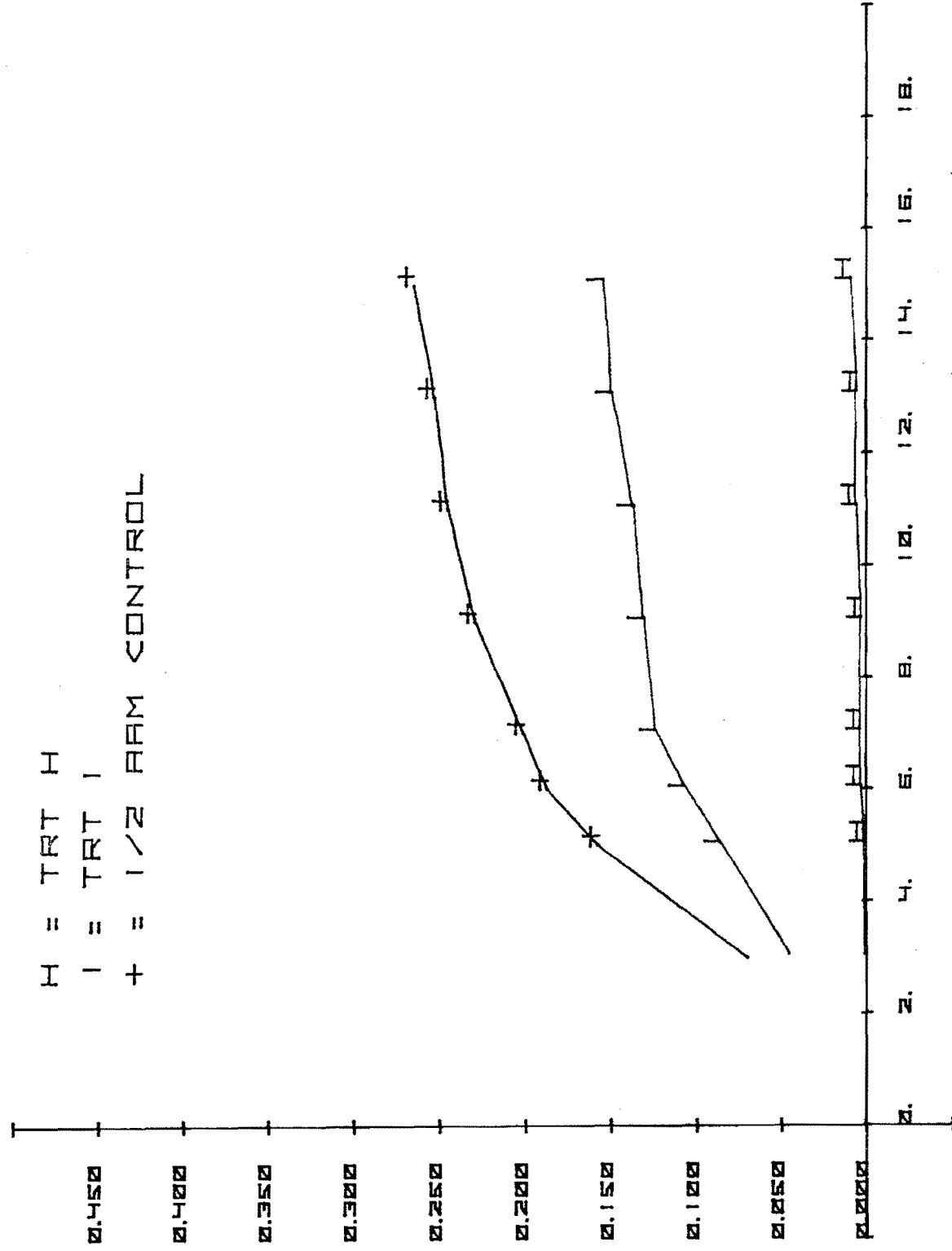
TIME [DAYS]

Figure 45.

LA PLATA AT COLO./N.M. BORDER
JANUARY 9 1978

H = TRT H
I = TRT I
+ = 1/2 AM CONTROL

OPTICAL DENSITY [750 NM. 1 CM.]



TIME [DAYS]

Figure 46.

LA PLATA RIVER AT COLO./N.M. BORDER
JANUARY 9 1978

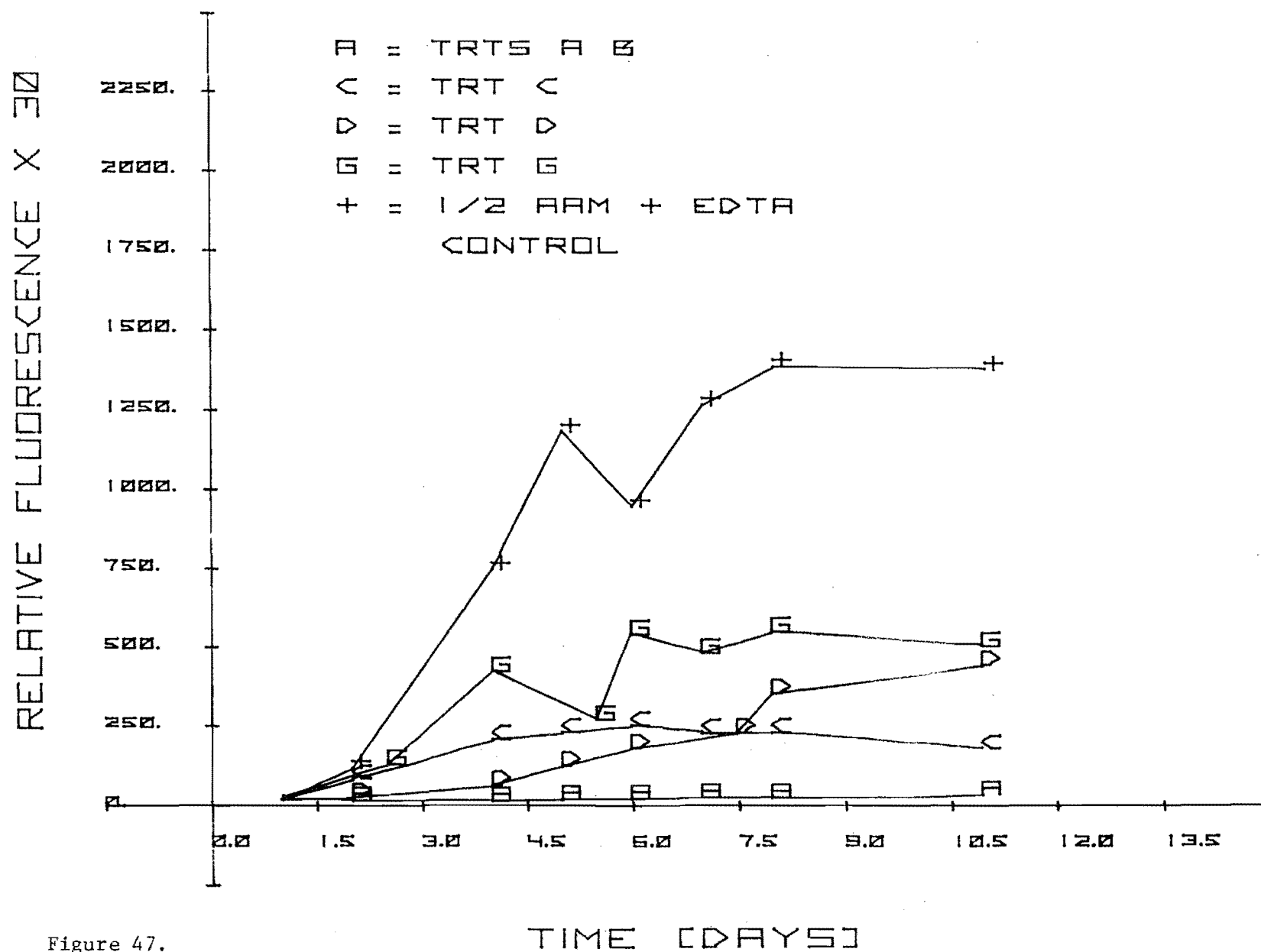


Figure 47.

LA PLATA RIVER AT COLO./N.M. BORDER

JANUARY 9 1978

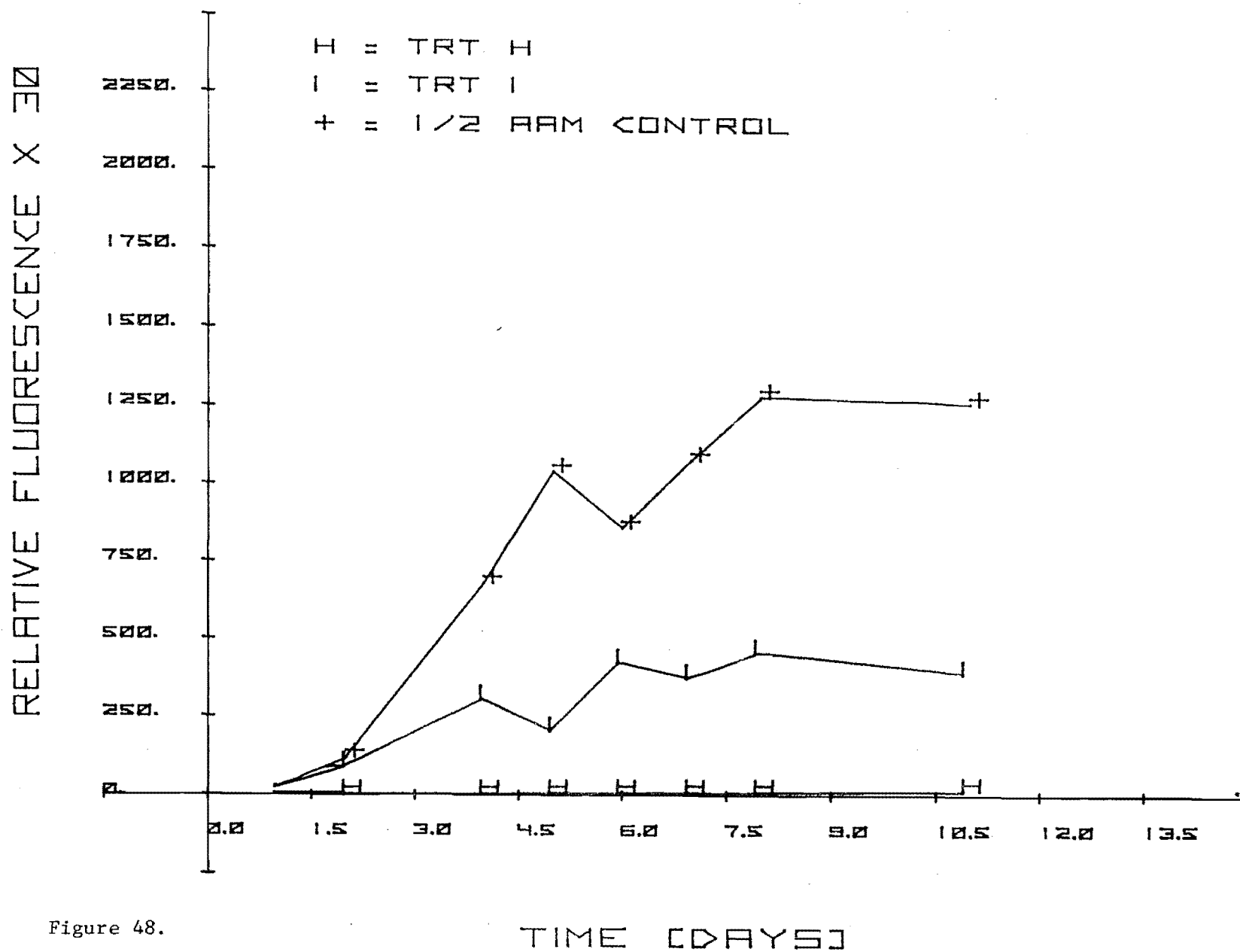


Figure 48.

LA PLATA RIVER AT COLO./N.M. BORDER
MARCH 8 1978 .

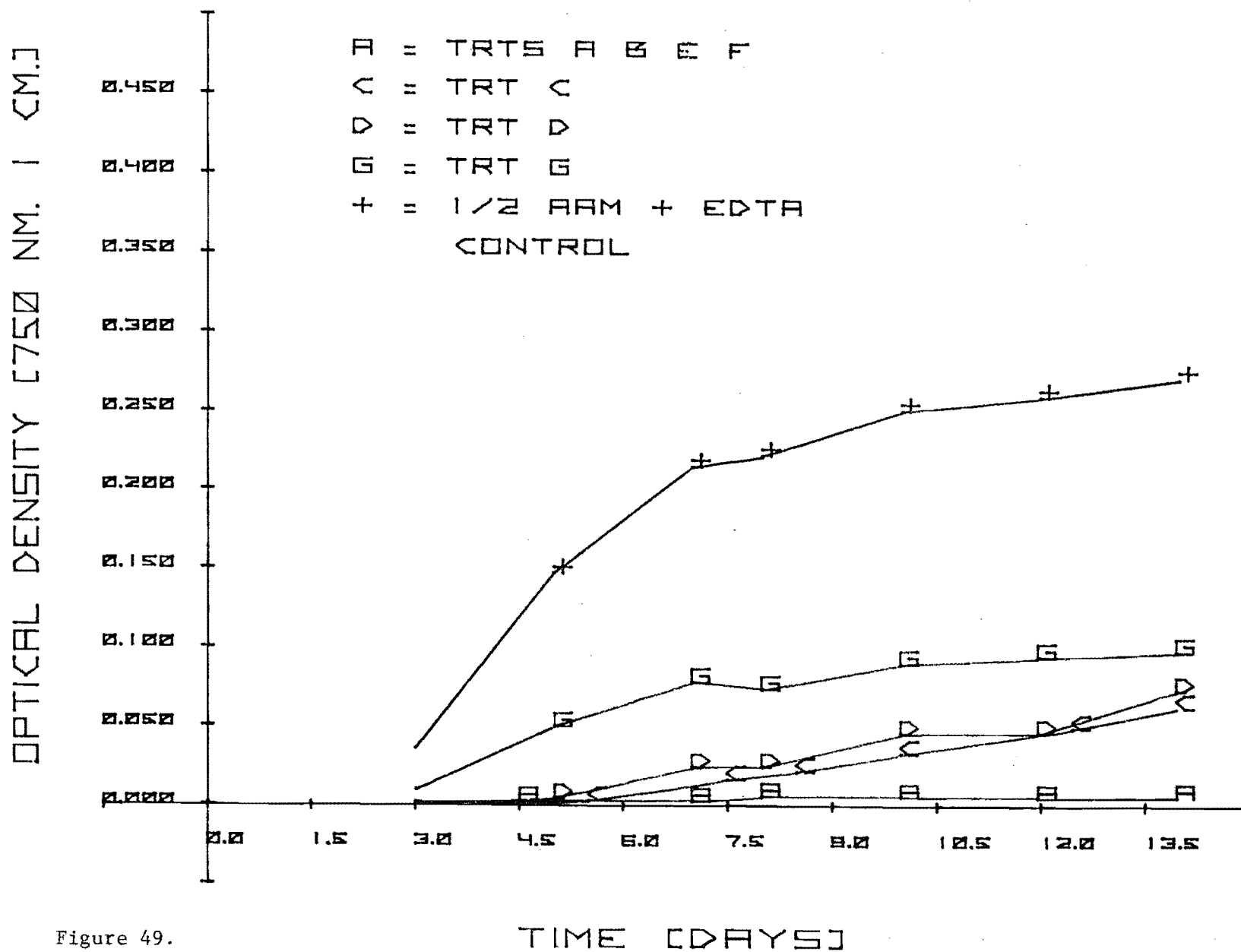
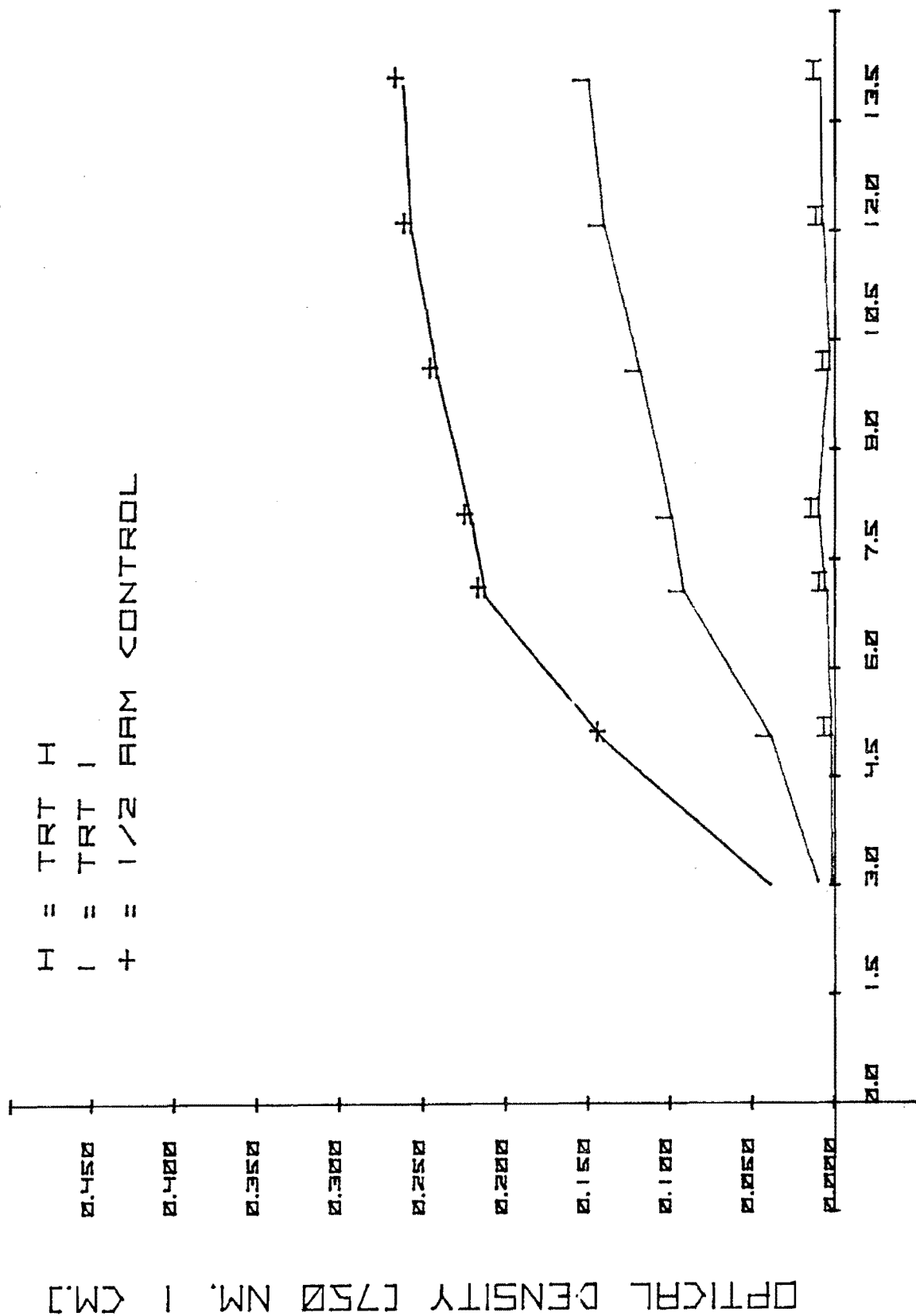


Figure 49.

LA PLATA RIVER AT COLD./N.M. BORDER
MARCH 8 1978

H = TRT H
I = TRT I
+ = 1/2 AM CONTROL



TIME [DAYS]

Figure 50.

LA PLATA RIVER AT COLO./N.M. BORDER
MARCH 8 1978

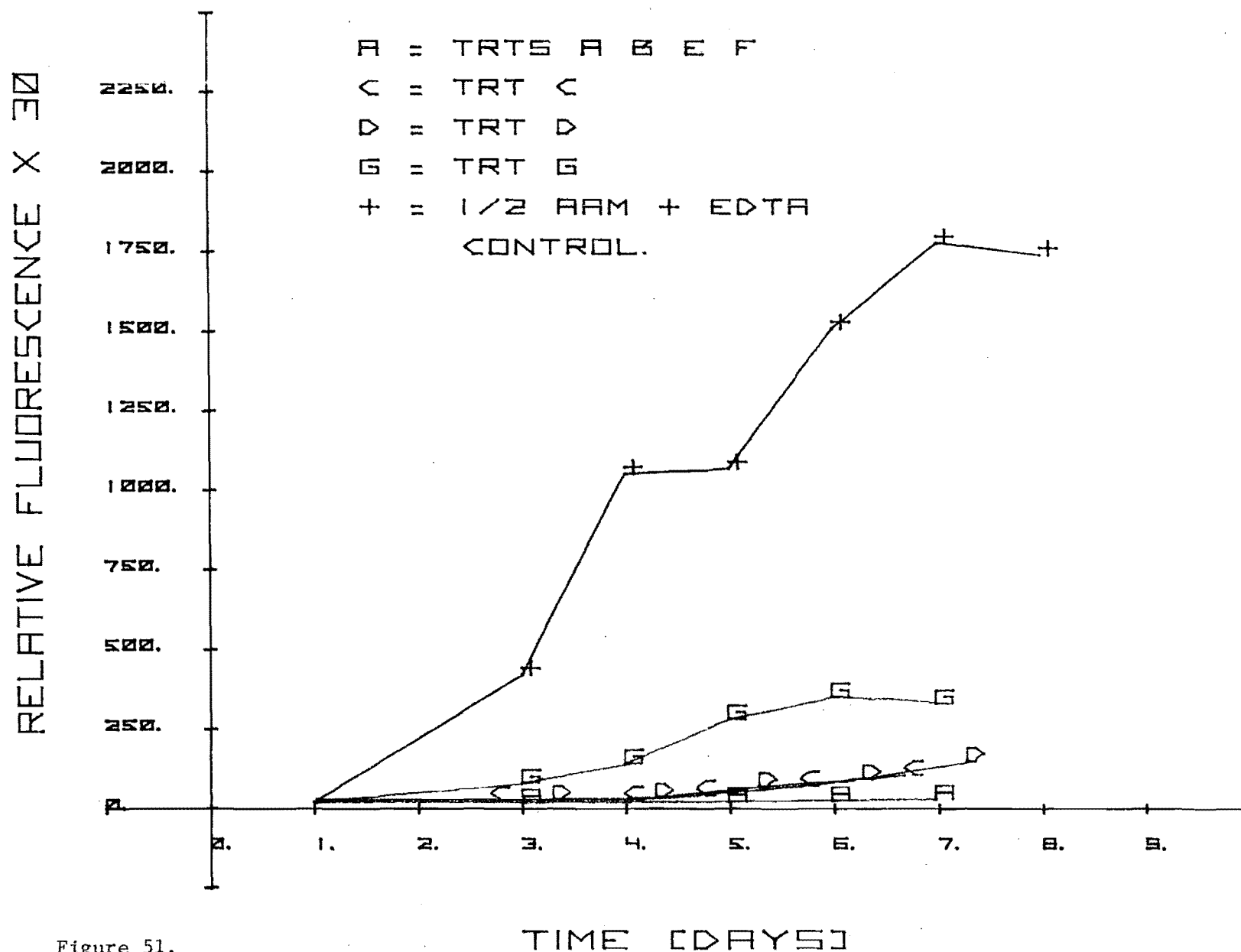


Figure 51.

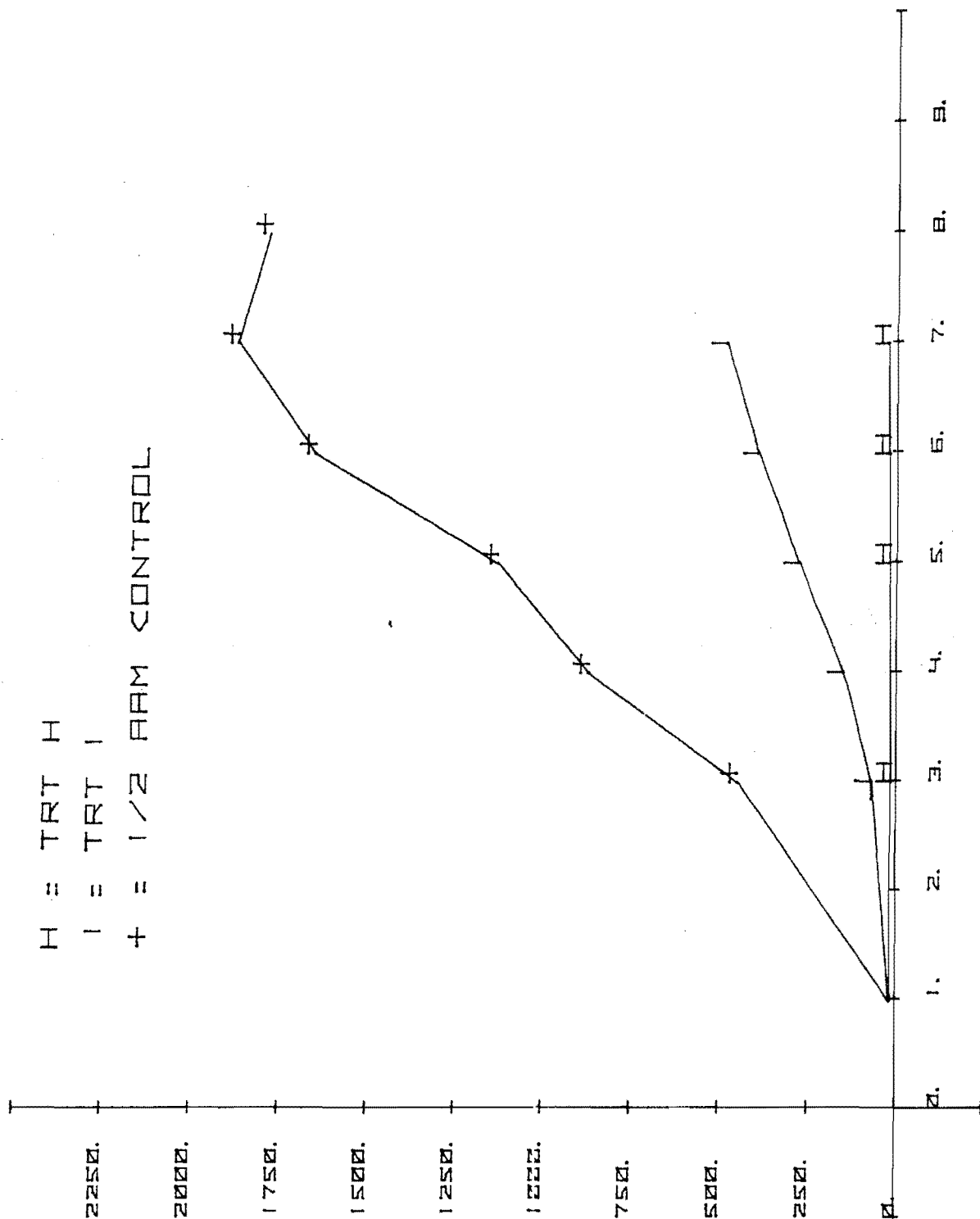
LA PLATA RIVER AT COLD./N.M. BORDER
MARCH 8 1978.

H = TRT H

I = TRT I

+ = 1/2 AM CONTROL

RELATIVE FLUORESCENCE X 10³



TIME [DAYS]

Figure 52.

LA PLATA RIVER AT COLD./N.M. BORDER
MAY 10, 1978

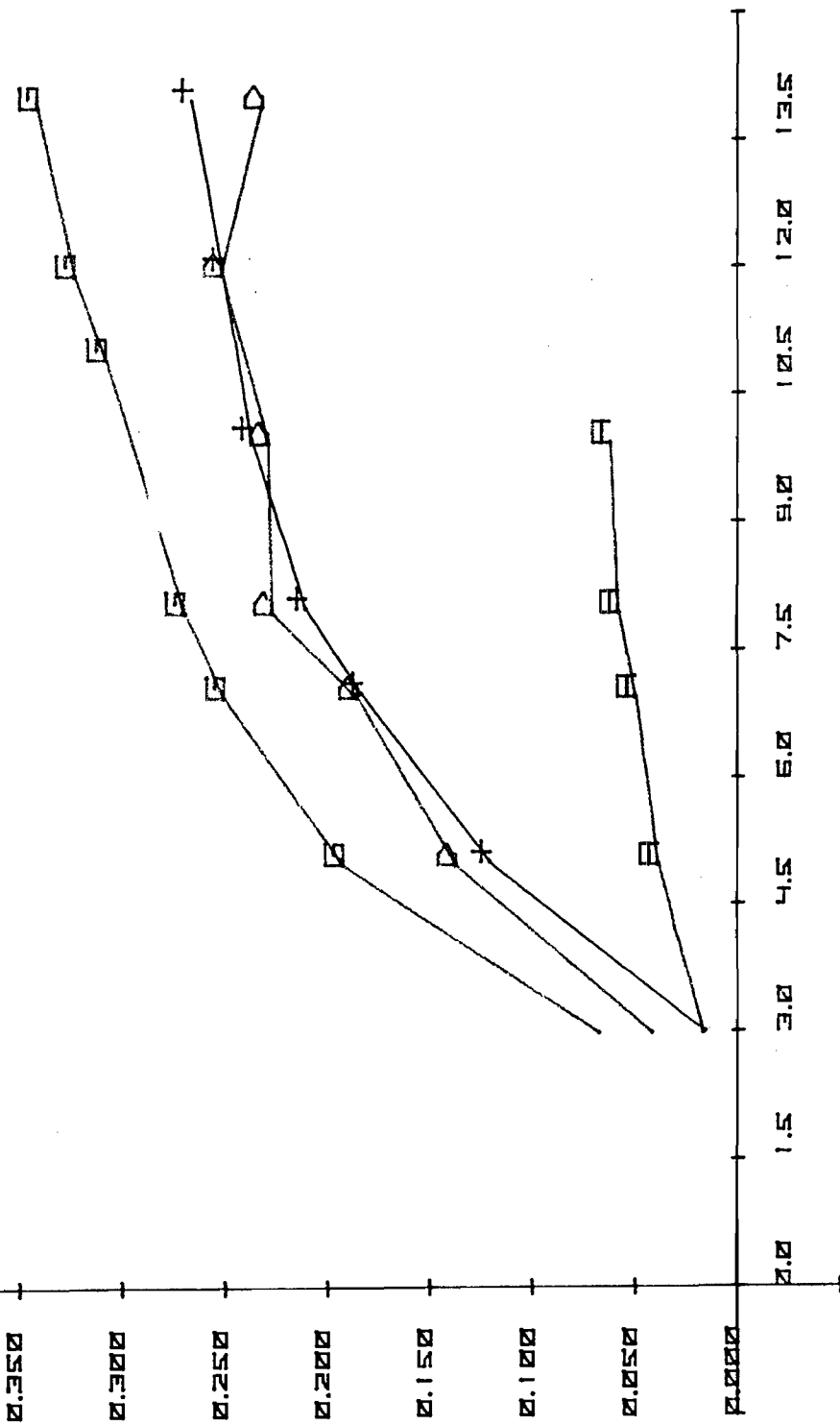
OPTICAL DENSITY, [750 NM. 1 CM.]

A = TRTS A, B, C, E, F

D = TRT D

G = TRT G

+ = 1/2 AM CONTROL



TIME (DAYS)

Figure 53.

LA PLATA RIVER AT COLO./N.M. BORDER
MAY 10, 1978

RELATIVE FLUORESCENCE X 30

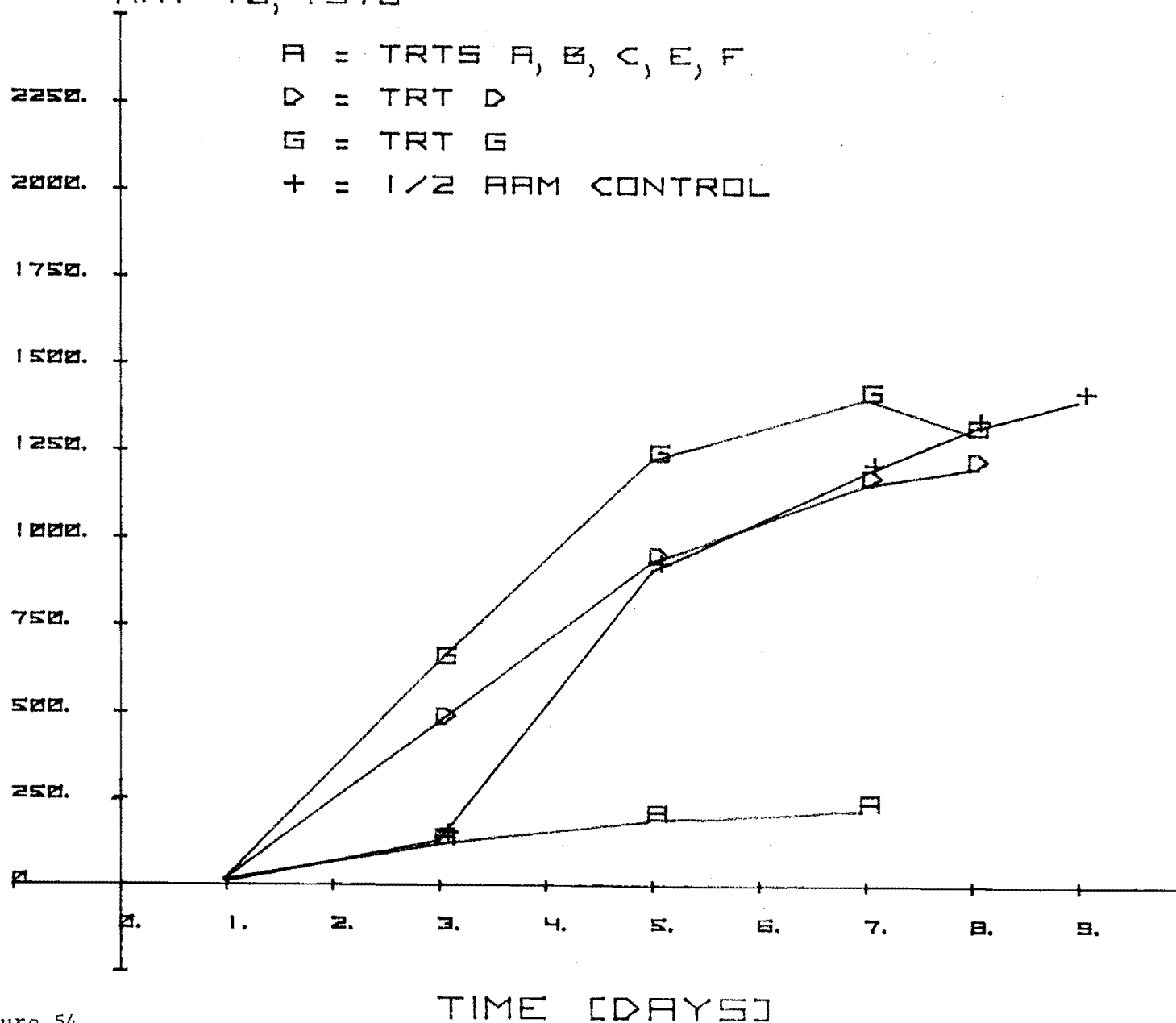


Figure 54.

Conclusions

1. Animas River at Durango

- a. A large concentration of heavy metals (possibly zinc) resulted in an infertile sample.
- b. The sample was limited by both phosphorus and nitrogen, after heavy metals were made unavailable using EDTA, indicating infertility even when toxicity was removed.
- c. There was a good correlation between chemical analysis and algal bioassay.
- d. This sample represents a non-productive body of water with oligotrophic to mesotrophic tendencies in the future.

2. Animas River at 32nd St. Bridge

- a. Heavy metal toxicity was indicated at this site.
- b. The sample was limited by both nitrogen and phosphorus during each sampling period with phosphorus being the most limiting.
- c. Algal bioassay confirmed the chemical analysis.
- d. An oligotrophic to mesotrophic condition can be expected at this site.

3. La Plata River at Colorado/New Mexico Border

- a. Hardness, calcium hardness specifically, seemed the most likely cause of lower than normal productivity.
- b. Phosphorus was the limiting nutrient in all bioassay but nitrogen became limiting as well.
- c. Chemical analyses and bioassay correlated well.
- d. The sample was classified as mesotrophic even though productivity increased slightly during the spring. The increased productivity in May, 1978 was not substantial enough to classify the sample as becoming eutrophic.

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