BIODEGRADATION (301E)

TEST SUBSTANCE

Identity: A mixture containing perfluorooctanesulfonate, which may also be referred to as PFOS, FC-95, or as a component of FC-203A or of FC-203 NFP. (1-Octanesulfonic acid) (CAS # 2795-39-3).

Remarks: The 3M production lot number was not noted. The test sample is FC-203 NFP Current information indicates it is a mixture of 1.34% PFOS, 25% diethylene glycol butyl ether, 67.85% water, 2.66 % Sultone foamer, 3% sodium octyl sulfate, 0.1% sodium lauryl sulfate, and 0.05% tolyltriazole.

The following summary applies to a mixture with incompletely characterized concentrations of impurities. Data may not accurately reflect degradation potential of the fluorochemical component of the test sample.

METHOD

Method: Modified OECD Screening Test, OECD 301E, with DOC Analysis, 1981 version.
Test type: Ready Biodegradability
GLP: No
Year Completed: 1982
Analytical monitoring: Dissolved organic carbon (DOC)
Statistical methods: Results were determined by calculation of the % DOC removal and graphic interpretation.
Test organism source: A 50:50 mix of soil extract and secondary effluent. The secondary effluent was the supernatant from an activated sludge aeration basin at the Metro Wastewater Treatment Plant, St. Paul, MN, while the soil was from the City of White Bear Lake, Ramsey County, MN.
Test condition:

Dilution water: Deionized water
Mineral Nutrient Medium: Nutrient medium per OECD 301E method (1981). Initial pH 7.0.
Reference and test solution preparation: The test material was prepared by dissolving 153 mg in two liters of mineral nutrient medium. This solution gives a final test concentration of 20 mg DOC/L. The reference substance, sodium benzoate, was prepared by dissolving 30.3 mg in one liter of mineral nutrient medium. This solution gives a final test concentration of 20 mg DOC/L.
Test vessels: Not given.
Incubation conditions: Constant dark conditions
Temperature: 23.5 – 24°C
Agitation: Continuously
Number of concentrations: 1 plus sodium benzoate (reference substance) and blank, all in duplicate.

Inoculum condition on test initiation: Not given .
Element Basis: Decrease in dissolved organic carbon compared to the blanks.
Test substance flask conditions: Not given.

RESULTS

Nominal concentrations: Blank control, sodium benzoate at ~20 mg DOC/L and at ~20 mg DOC/L plus $HgCL_2$ (inhibited), test material at ~20 mg DOC/L and at ~20 mg DOC/L plus $HgCl_2$, all in duplicate.

Element values: 27-day Degradation = 76.7% duplicate 1 and 81.3% duplicate 2. Mean value = 79%

Remarks: Testing was conducted on the mixture as described in the Test Substance Remarks field. The values reported apply to that mixture and not the fluorochemical proportion alone.

CONCLUSIONS

The FC-203 NFP % degradation based on the mean DOC removal value was 79 after 27 days.

Submitter: 3M Company, Environmental Laboratory, P.O. Box 33331, St. Paul, Minnesota, 55133

DATA QUALITY

Reliability: Klimisch ranking = 2. This study meets the criteria for quality testing. However, the sample purity was not properly characterized and the study lacks analytical confirmation of the amount of fluorochemical proportion in the solution.

REFERENCES

The studies were conducted by the 3M Company, Environmental Laboratory, St. Paul, MN, Lab Request number 8483, 1982.

OTHER

Last changed: 6/28/00

3M COMPANY
Form 6747-11-A

TECHNICAL REPORT SUMMARY

Date: 6/30/80

TO: TECHNICAL COMMUNICATIONS CENTER – 201-2CN

(Important – If report is printed on both sides of paper, send two copies to TCC.)

Division: Environmental Laboratory (EE & PC)	Dept. Number: 0535
Project: Commercial Chemicals Division	Project Number: 9970012600
Report Title: Biodegradation of "LIGHT WATER" Products in OECD Test-6/80	Report Number: 040
To: John A. Pignato - Commercial Chemicals Division - 236-2A (01)	
Author(s): Eric A. Reiner - Environmental Lab (EE&PC) - 21-BW (63)	Employee Number(s): 47816
Notebook Reference: None - See 3M Environmental Lab Request No. 5541S	No. of Pages Including Coversheet: 31

SECURITY ▶ ☒ Open ☐ Closed | 3M CHEMICAL REGISTRY ▶ | New Chemicals Reported ☐ Yes ☒ No

KEYWORDS: (Select terms from 3M Thesaurus. Suggest other applicable terms.)

Environmental Laboratory (EE & PC)

AFFF

TOC

CURRENT OBJECTIVE:

To use the internationally recognized OECD Screening Test with DOC analysis to show that "LIGHT WATER" products are highly biodegradable.

REPORT ABSTRACT: (200-250 words) This abstract information is distributed by the Technical Communications Center to alert 3M'ers to Company R&D.

This study used the "Modified OECD Screening Test with DOC Analysis" and supplemental parallel sterile controls to conclusively demonstrate the extensive biodegradability of "LIGHT WATER" Brand AFFF products: FC-203, FC-206, and FC-3017. In 14 days, the dissolved organic carbon (DOC) levels of FC-206 degraded by 90% and FC-3017 was 94% degraded. FC-203 showed 93% DOC degradation in 21 days. The parallel sterile controls proved that this DOC loss was not due to chemical or physical processes such as adsorption, volatilization, or precipitation of the parent material.

Information Liaison Initials: MO

BIODEGRADATION OF "LIGHT WATER" PRODUCTS IN OECD TEST

INTRODUCTION

The 3M Environmental Laboratory chose to attempt to demonstrate the biodegradability of "LIGHT WATER" products using the OECD screening method because the test has wide international acceptance, and the Environmental Lab has had experience using the OECD test as a participant in an international ring test. These results will complement the existing BOD data that show these products to have a high degree of biodegradability.

The OECD test is run at 20-25°C and it measures only dissolved organic carbon. For these water-miscible "LIGHT WATER" products, which are not expected to volatilize, precipitate, or be adsorbed during testing, the OECD test is an appropriate method for measuring % total degradation. The TOC analysis method even measures the perfluorinated portion of these products.

METHODS AND MATERIALS

Methods and Conditions - Wade A. Scheil performed this testing following the September 19, 1979, version of the Modified OECD Screening Test with DOC Analysis(1). A copy of this method is attached (Appendix 1).

Reductions in the scale of the test were necessary to adapt it to available shaking equipment. Five hundred-ml Erlenmeyer flasks replaced 2-liter flasks, 250 ml of inoculated nutrient solution replaced 900 ml, and the sample size was 25, not 30 ml. As in the prescribed procedure, the analyst used only the last 10 ml of sample filtrate for DOC analysis.

A calibrated chart recorder monitored the temperature which stayed within the 20-25°C range during the 28-day test, except for a 3-4 hr. period when it reached 25.5°C. The temperature was most frequently near the high end of the range (23.5-24°C).

Chemicals - All chemicals were reagent grade unless otherwise noted.

The 3M "LIGHT WATER" products used were samples of commercial material of Antwerp manufacture. Their label identified them as FC-203 NFP, FC-3017 Lot 2303, and FC-206 (FM 3824) Lot 2330 10/79.

The hydroquinone used as the calibration compound was Aldrich reagent grade hydroquinone 98.5%, Lot 100147. Four chemical supply houses were contacted in an attempt to obtain the 99.5% pure hydroquinone prescribed by the Modified OECD Screening Test, but this purity is apparenty not readiy available in the USA. We expect that this reduced purity will have no signficant effect on results. The impurities have no apparent inhibitory effect on microorganisms and their presence could make only a trivial change in TOC measurements.

Water used in the preparation of all solutions, except the inoculum, was St. Paul city water passed through a carbon bed, macroporous anionic resin bed, and 2 mixed bed deionizers, and then filtered through a 0.2-um filter. This water was also used to make up losses to evaporation. Stocks of water collected at the start of the experiment ensured consistent water quality.

In preparing nutrient solutions, 26.4 g of anhydrous Na_2HPO_4 replaced the prescribed 33.4 g of $NH_2HPO_4 \cdot 2H_2O$, 36.3 g of $CaCl_2 \cdot 2H_2O$ replaced 27.5 g $CaCl_2$. These replacements yielded solutions and concentrations identical to those required by the OECD Method. The optional yeast extract solution was used in place of the vitamin solution (1.6.1.2(f)). The 100 mg of Fe-chelate used in the trace element solution consisted of equimolar amounts of $FeCl_3$ and EDTA (30 mg $FeCl_3$ and 70 mg EDTA).

Test Concentrations - DOC measurements of sample stock dilutions served as the basis for adjusting initial test concentrations to approximately 40 mg of carbon per liter. This is the highest concentration in the range recommended by the OECD test. Use of this high initial concentration increased the sensitivity of % DOC removal measurements. The low toxicity of "LIGHT WATER" products to microorganisms made significant inhibition at this concentration unlikely.

Adjustment of hydroquinone, the calibration compound, to 20 mg of carbon per liter was done by weight based on its known carbon concentration of 65.4%.

Two blank controls, instead of 1, were utilized in making blank DOC corrections.

Inoculum - The inoculum was 50-50 mixture prepared according to steps 1.6.2.1 and 1.6.2.2 of the OECD test using fresh soil from E. A. Reiner's garden and the supernatant of a sample from an activated sludge aeration basin at the St. Paul Metro Plant. The filter paper used was Whatman^R 54, and the water for soil extraction was chlorine-free well water.

Sterile Controls - Replicate vessels containing 400 mg/l $HgCl_2$ served as sterile controls for each test material and the calibration compound. Handling of these sterile controls was identical to the handling of the viable cultures. They had the same innoculum and sampling frequency, but the analyst only made DOC measurements on the 0- and 28-day samples.

Sample Preservation - The sample preservation method used was that prescribed by the TOC manufacturer[4]. The method involves adding 1 drop of concentrated HCl to the 10 ml filtered samples, bringing the sample to <pH 2, and storing the samples under refrigeration in vials with aluminum foil-lined caps. The method is an effective preservation method. The manufacturer indicates that the method can stabilize calibration solutions made from potassium hydrogen phthalate, a readily biodegradable material, for several months.

Since the acidification in this preservation method was also a necessary step in the TOC analysis protocol, its use was the most practical and allowed elimination of the $HgCl_2$ preservation technique described in the OECD method. All TOC analyses were made within a month of the preservation of the samples.

Instruments and equipment - The organic carbon analyzer used was a Dohrman DC-52A. Its sensitivity limit is between 1 and 2 mg of carbon per liter.

The stoppers for the reaction flasks were clean, porous, plastic foam plugs. Gelman 0.2 um membrane filters, (Part No. 64814) boiled 3 times and stored in deionized water, served to filter samples for DOC analysis.

Glassware cleaning involved soaking in chromic acid cleaning solution (Chromerge^R) followed by 6 deionized water rinses.

RESULTS AND DISCUSSION

Table 1 summarizes the results of this study. Appendices 1 and 2 contain the actual data sheets and plots of the degradation as a function of time. All 3 "LIGHT WATER" AFFF products degraded nearly completely.

TABLE 1
Percent Degradation of Hydroquinone and "LIGHT WATER" AFFF Products FC-203, FC-206, and FC-3017 With Time in Modified OECD Screening Test

	% Degradation at Day:				
Product	7	14	21	27	28
FC-203	13	85	93	91	93
FC-206	23	90	94	93	92
FC-3017	37	94	96	96	95
Hydro-quinone	89	92	97	94	93

In the case of the highly water soluble "LIGHT WATER" products, it is very unlikely that physical means such as adsorption, volatilization, or precipitation caused the loss of soluble TOC (DOC). This is substantiated by the relatively high oxygen demand observed in BOD tests of the 3M Environmental Laboratory (FC-203 $BOD_5/COD = 0.5$, FC-206 $BOD_5/COD = 0.5$).

The sterile control data in Table 2 provide further substantial evidence that the soluble TOC loss from the test samples containing "LIGHT WATER" products is not due to adsorption, volatilization, or precipitation. These samples were handled identically to the test samples except that they contained $HgCl_2$ to prevent microbial growth.

At the end of the 28-day test period, they still contained nearly all the initial DOC. Thus loss of soluble TOC by these physical modes was not a major factor, at least for nonmetabolized "LIGHT WATER" components. This control doesn't prove that physical processes did not remove "LIGHT WATER" metabolites in the test runs, since no metabolism occurred in these sterile controls, but the catabolic* formation of less water soluble materials is unlikely because catabolic products are usually more polar and smaller.

TABLE 2

DOC Loss from Sterile Controls

Product	% DOC Remaining at Day 28
FC-203	91.3
FC-206	94.2
FC-3017	88.1
Hydroquinone	77.5

An off-white precipitate formed in the hydroquinone sterile control on day 0 prior to filtering for DOC analysis. The precipitate was slightly brownish, as was the solution in the hydroquinone sterile control after 21 days. This suggests that hydroquinone is not stable, at least in the presence of $HgCl_2$, throughout the course of the 28-day OECD experiment. This apparent tendency of hydroquinone to spontaneously form insoluble humus-like material makes it an inappropriate control compound for the OECD test. Its partial precipitation with $HgCl_2$ also casts some doubts about the appropriateness of using the $HgCl_2$ preservation procedure in the OECD method.

CONCLUSION

The present study conclusively demonstrates that "LIGHT WATER" products, FC-203, FC-206, and FC-3017, are nearly completely degraded in 21 days under the conditions of the modified OECD Screening Test.

REFERENCES

(1) Organization for Economic Co-operation and Development Chemicals Testing Program Expert Group on Degradation/Accumulation, Dec., 1979, Test Guideline for the Modified OECD Screening Test with DOC Analysis (Level I) (H. G. Nosler) Revision of Sept. 19, 1979.

(2) Dohrman DC-52A Operating Manual, 4th Ed., 1978, p. 3-1.

Appendices: 1 - OECD Test Guidelines (Sept. 19, 1979)
2 - Data Sheets
3 - Graphs

* Catabolism is biologically facilitated breakdown to less complex molecules.

APPENDIX 1

Tokyo, December 1st, 1979

C 118/79/Int.

OECD - Chemicals Testing Programme
Expert Group C, Degradation/Accumulation

Mbdified OECD Screening Test with DOC Analysis

Test Guideline C 118/79/Int.

Test Guideline for the Modified OECD Screening Test with DOC Analysis

Date of last revision: Sept. 19,1979

Level I test for ready biodegradability

1. Prerequisites

It has to be known whether the test material is soluble in the concentration range employed in the test (corresponding to 5 - 40 mg DOC/l).

2. Guidance Information

Knowledge of the bacterial toxicity or inhibitory properties of the test material is not unequivocally required but constitutes useful information for the conduction of the test.

3. Qualifying Statements

The method is suited for the measurement of the aerobic ultimate biodegradability of water soluble, non-volatile organic compounds. It is unsuited for the biodegradability evaluation of mixtures.

(b) 3

I S. 2 97 012/5

Sept. 19. 1979

Modified OECD Screening Test with DOC Analysis

Preamble

It has to be realized that the following procedure for the modified OECD Screening Test has to be regarded in some points as provisional since several important features such as the calibration compound and the new test duration of 28 days are completely untried yet. However, since the test constitutes a modification the official, widely practised and well accepted OECD Screening Test for the biodegradability evaluation of surfactants there should exist a good prospect for success.

September 19., 1979

OECD Chemicals Testing Programme

Test Guideline for the Modified
OECD Screening Test with DOC Analysis

0. The test procedure constitutes a modification of the OECD Screening Test (OECD Environment Directorate, Proposed Method for the Determination of the Biodegradability of Surfactants Used in Synthetic Detergents, Paris 1976, and council directive of Nov. 22, 1973, on the approximation of the laws of the member states relating to methods of testing the biodegradability of anionic surfactants (73/405/EEC), Official Journal of the European Communities No. 4 347/53 of Dec. 17, 1973) for the application of the dissolved organic carbon (DOC) analysis.

1. Method

1.1 Introduction: Purpose, scope, relevance, and application of test and explanation of limits.

The purpose of the method is the measurement of the ultimate biodegradability of water soluble, non - volatile organic compounds in an aerobic, aqueous medium at a starting test concentration corresponding to 5 - 40 mg DOC/l (In order to avoid inhibitory effects it is in the investigator`s own interest

to choose as low a starting concentration as his analytical capability permits)

1.2 Definitions and units

1.2.1 Definition of biodegradability

$$D_t = \left[1 - \frac{C_t - C_{bl_t}}{C_o - C_{bl_o}} \right] \times 100$$

where

D_t = degradation in percent DOC-removal at time t

C_o = starting DOC concentration of the culture medium (mg DOC/l)

C_t = DOC concentration of the culture medium at time t (mg DOC/l)

C_{bl_o} = starting DOC concentration of the blank (mg DOC/l)

C_{bl_t} = DOC concentration of the blank at time t (mg DOC/l)

1.2.2 Units

The degradation is stated as the percentage DOC-removal within 28 days with respect to the test material (% DOC-removal)

1.3 Reference compounds

1.3.1 Calibration compound

The calibration compound used in this test is hydroquinone at a concentration corresponding to 20 mg DOC/l. Hydroquinone has to exhibit a DOC-removal of ≧60 % within 28 days, otherwise the test is regarded as invalid.

1.4 The principle of the method

A predetermined amount of the compound is dissolved in an inorganic medium (mineral nutrient solution, fortified with a trace element and essential vitamin solution), providing a concentration corresponding to 5 - 40 mg DOC/l. The solution is inoculated with a small number of microorganisms from a mixed population and aerated at 293 - 298 (20 - 25°C) in the dark or at least in diffuse light only. The degradation is followed by DOC analysis over a 28 day period. The procedure is checked by means of a standard (hydroquinone).

A control with inoculation but without either test material or standard is run parallel for the determination of DOC blanks.

1.5 Quality criteria

1.5.1 Reproducibility

The reproducibility of the method is appropriate for a screening test which has solely an acceptance but no rejective function.

1.5.2 Sensitivity

The sensitivity of the method is largely determined by the sensitivity limit of the organic carbon analysis which is 0.5 mg C/l at the present state of the art.

1.5.3 Specificity, applicability

Applicable for the biodegradability evaluation of water soluble, non - volatile organic compounds.

1.5.4 Possibility of standardization

The test version with specific analyses for anionic and nonionic surfactants is standardized as "OECD Screening Test".

1.5.5 Possibility of automation

Parts of the test, e.g., the analysis,can be automated, although hardly the total procedure. The procedure is, though, well suited for being operated with whole series of test materials.

1.5.6 Costs (in 1978 Swiss Francs)

1.5.6.1 Equipment

Glassware	2000,-
shaking machine	6000 - 20000,-
Carbon analyzer	42000,-
Miscellaneous (pH meter, balance, provision for air donditioning)	5000,-

1.5.6.2	Person - hours	10	
1.5.6.3	Approximate total cost per test (investments assumed to be amortized)	750,-	- 1000,-

1.6 Description of the method

1.6.1 Reagents and materials

1.6.1.1 Deionized water

Deionized or distilled water free of toxic substances (copper in particular), for general use as a solvent. Water which has been deionized by distillation or ion exchange is suitable.

A high purity of this test water is necessary in view of the DOC analyses in the concentration range of 0 - 40 mg/l. The contaminations result from inherent impurities but also from the ion exchange resins and microbial developments (bacteria, algae under the influence of light etc). Only one water charge must be used for one test series which is to be controlled beforehand by DOC analysis. If necessary, suitable water may be gained by UV irradiation or other means.

1.6.1.2 Nutrient solution

Mix 1 ml each of the following solutions (a) to (f) and make up to a volume of 1 l with water 1.6.1.1

(a) KH_2PO_4 A.R. 8.5 g
K_2HPO_4 A.R. 21.75g
$Na_2HPO_4 . 2 H_2O$ A.R. 33.4 g
NH_4Cl A.R. 20.0 g
in 1000 ml of water 1.6.1.1
the pH value should be 7.2

(b) 22.5 g of $MgSO_4 . 7 H_2O$ A.R. dissolved in 1000 ml of water (3.3.1)

(c) 27.5 g of $CaCl_2$ A.R. dissolved in 1000 ml of water (3.3.1)

(d) 0.25 g of $FeCl_3 . 6H_2O$ A.R. dissolved in 1000 ml of water (3.3.1)
This solution is prepared freshly immediately before use.

(e) Trace element solution

$MnSO_4 . 4 H_2O$	39.9 mg (30.23 mg $MnSO_4 . H_2O$)
H_3BO_3	57,2 mg
$ZnSO_4 . 7 H_2O$	42.8 mg
$(NH_4)_6Mo_7O_{24}$	34.7 mg (36.85 mg $(NH_4)_6Mo_7O_{24}.4H_2O$)

Fe - chelate ($FeCl_3$,EDTA)	100 mg
water 1.6.1.1	1000 ml

Sterilisation of the trace element stock solution at 393 (K) (120°C), 2 atm., 20 min.

(f) Vitamin solution

Biotin	0.2 mg
Nicotinic acid	2.0 mg
Thiamine	1.0 mg
p-Aminobenzoic acid	1.0 mg
Pantothenic acid	1.0 mg
Pyridoxamine	5.0 mg
Cyanocobalamine	2.0 mg
Folic acid	5.0 mg
water 1.6.1.1	100 ml

The solution is filtered sterile (0.2 µm). Instead of solution 1.6.1.2 (f) 15 mg of yeast extraxt may be used per 100 ml of water 1.6.1.1.

1.6.1.3 Biodegradability standard
Hydroquinone 99,5 % DAB Erg. B. 6

1.6.1.4 Mercuric chloride solution
1 per cent of $HgCl_2$ in water

1.6.1.5 Shaking machine accomodating 2 ltr. Erlenmeyer flasks either with automatic temperature control or used in a constant temperature room at 293 - 298(K) (20 - 25°C)

1.6.1.6 Narrow neck - 2 ltr. Erlenmeyer flasks. (Creased fluted flasks are recommended) The flasks must be carefully cleaned with, e.g., alcoholic hydrochloric

e use, rinsed and dried in order to avoid
nation with residues from previous tests.
lasks also have to be cleaned before their
rst use since they may be contaminated.

1.6.1.7 Membrane filtration apparatus

1.6.1.8 Membrane filters 0,2 µm

1.6.1.9 Carbon analyzer

1.6.2 Inoculation

Either of the following three alternatives may be used as inoculum or a composite sample thereof.

1.6.2.1 Inoculum from secondary effluent

The inoculum is gained preferentially from a secondary effluent of good quality collected from a treatment plant dealing with a predominantly domestic sewage. The effluent must be kept under aerobic conditions in the period between sampling and use. To prepare the inoculum the sample is filtered through a coarse filter, the first 200 ml being discarded. The filtrate is kept aerobic until used. The inoculum must be used on the day of collection.

1.6.2.2 Inoculum from soil

100 g of soil (fertile, not sterile) are suspended in 1000 ml of chlorine-free drinking water (soils with an extremely large content of clay, sand or organic carbon are unsuited). After stirring the suspension is allowed to settle for 30 minutes.

The supernatant is filtered through a coarse filter paper, the first 200 ml being discarded. The filtrate is aerated immediately and until use. The inoculum must be used on the day of collection.

1.6.2.3 Inoculum from a surface water

An inoculum is drawn from a suitable surface water. The sample is filtered through a coarse paper, the first 2oo ml being discarded. The filtrate is kept aerobic until used. The inoculum must be used on the day of collection.

1.6.2.4 Composite inoculum

Equal volumes of the 3 inoculum samples are united, mixed well, and the final inoculum drawn from this mixture.

The suitability of the inoculum is checked by means of the standard hydroquinone.

1.6.3 Conduction of the test

1.6.3.1 Procedure

The test materials are evaluated simultaneously in duplicates together with the standard (1.6.1.3) and a control test with inoculation but without either test or standard material for the determination of DOC blanks.

The standard material has to attain ≥ 60 % DOC removal within 28 days at a starting concentration corresponding to 20 mg DOC/1. If < 60 % DOC removal are achieved the whole series has to be discarded +)

+) This limit is based on present experience with the 19 day version of the test. A revision of this limit or even of the standard might have to be considered after the accumulation of experience with the new 28 day version of the test.

A stock solution of the test material in water (1.6.1.1) is prepared. So much of this stock solution is added to the nutrient solution (1.6.1.2) that a carbon concentration of 5 - 40 mg DOC/l is attained. The starting concentration of the standard hydroquinone is, though, 20 mg DOC/l.

Two reaction vessels (1.6.1.5) are each filled with 900 ml of the nutrient solution and inoculated with 0,5 ml/l of the inoculum (1.6.2). The opening of the vessel is covered with, e.g., aluminum foil in such a way that the exchange of air between the flask and the surrounding atmosphere is not unduly impeded (Cotton wool is unsuited because of the DOC analysis). The vessels are then inserted in the shaking machine. The temperature of 293 -298 K (20 -25 °C) must be maintained unchanged during the test, and the vessels should be shielded from light. The air should be free of pollutants and toxic materials (chlorinated solvents etc).

In the course of the biodegradation test the DOC concentrations are determined in duplicate (1.6.4.2) at the beginning (day 0), and on the 27. and 28. day. Three additional analyses have to be performed

in rather regular time intervals (~7., ~14., and ~21. day)

The analyses are registered in the attached form sheet and evaluated.

Only the necessary volumes of culture medium may be drawn for each determination; however, they have to be large enough for the membrane filtration or centrifugation preceeding the carbon determination. The latter requires differing volumes for the different instruments. Evaporation losses of the culture medium are to be made up by adding water (1.6.1.1) in the required amounts. The culture medium is to be mixed well before withdrawing a sample. Material adhering to the wall of the vessel has to be dissolved or suspended before sampling. The membrane filtration or centrifugation has to be done immediately. The filtered or centrifuged samples have to be analyzed on the same day, otherwise they must be preserved with 0,05 ml of the $HgCl_2$ solution (1.6.1.4) for each 10ml of nutrient medium or by storing them at 2 - 4°C. The biodegradability test is valid provided the standard exhibits a degradation rate within the specified range. The test can be finished before the 28. day if complete mineralization is accomplished.

All steps require great care and cleanliness of the vessels, pipettes etc. but not sterility.

1.6.3.2 Calculation of results

The degradation at the time t is calculated from the determinations of the DOC concentrations at the beginning (C_o) and the time t (C_t) according to

$$D_t = \left[1 - \frac{C_t - C_{bl_t}}{C_o - C_{bl_o}} \right] \times 100$$

where

D_t = degradation in per cent at time t

C_o = measured starting DOC concentration of the inoculated culture medium (mg DOC/l)

C_t = DOC concentration of the culture medium at time t (mg DOC/l)

C_{bl_o} = starting DOC blank of the mineral nutrient solution with inoculation but without test material (mg DOC/l)

C_{bl_t} = DOC blank of the mineral nutrient solution with inoculation but without test material at the time t (mg DOC/l)

The degradation rates are calculated to the nearest 0,1 %. The means of the D_t values are calculated and reported to the nearest full per cent. Results

ending in 0,5 are rounded up to the nearest whole number. The course of the degradation test is followed graphically in a diagram as shown in the attached example. The results are reported on the attached data sheet.

The results of the degradation test are valid if the condition is met that in the same test series the standard yields ≧ 60 % DOC-removal.

1.6.4 Analytical means

1.6.4.1 Membrane filter 0,2 µm, 25 mm Ø (1.6.1.6). Preparation of the filters: membrane filters are impregnated with surfactants for hydrophilization. Thus each filter contains up to several mg of soluble carbon which would interfere in the biodegradability determinations. Therefore the filters are purified from surfactants and other soluble organic interferences by boiling them 3 times 1 hr each in deionized water. These filters may be stored in water (1.6.1.1) for at least one week.
Other membrane filters are suitable if it is assured that they neither release carbon nor adsorb the compound in the filtration step.

If the samples are centrifuged, this has to be done at 40 000 m sec^{-2} (4000 g) for 15 minutes, preferably in a refrigerated centrifuge, in any case $< 40^{\circ}C$.

(Remark: the differentiation TOC:DOC by centrifugation at very low concentrations does not seem to work well since either not all bacteria are removed or carbon as part of the bacterial plasma is redissolved. At higher test concentrations (⩾10 mg C/l) and the same small inoculation the centrifugation error seems to be comparatively small).

Form Sheet for the Modified OECD Screening Test

Exp. no.: _____

Date of start of test: _____

Test / standard material: _____

Theoretical test conc.: _____ mg DOC/l

Inoculum: _____

Carbon analyzer: _____

A: Controls:

	TOC* mg/l	DOC** mg/l
Stock solution of the test material (1000 mg/l, dilution .../1000 ml of nutrient solution)		

B: Carbon determinations:

Culture medium	Flask no.	Analyses	Theor. conc. mg/l	DOC - concentrations after x days mg/l: 0 (C_0)	~7	~14	~21	27	28
Mineral nutrient solution with test material and with inoculum	1	a_1	/						
		a_2	/						
		$m_1 = \frac{a_1 + a_2}{2}$ (C_t)							
	2	b_1	/						
		b_2	/						
		$m_2 = \frac{b_1 + b_2}{2}$ (C_t)							
Mineral nutrient solution without test material but with inoculum	Blank	c_1	/						
		c_2	/						
		$m_3 = \frac{c_1 + c_2}{2}$ (C_{b1})	/						

C: Evaluation of raw data:

	Flask no.		% DOC removal after x days: 7	14	21	27	28
DOC - concentrations minus blanks	1	$D_1 = \frac{m_1 - m_3}{m_1} \cdot 100$					
	2	$D_2 = \frac{m_2 - m_3}{m_2} \cdot 100$					
	mean	$D = \frac{D_1 + D_2}{2}$ · for day x					

1.6.4.2 The DOC measurement

The sample withdrawn from the culture medium (about 30 ml) is centrifuged or membrane filtered immediately in the filtration apparatus (1.6.1.6) using the membrane filters prepared acc. to 1.6.4.1. The first 20 ml of the filtrate are discarded.

The DOC concentration is determined twice in the remaining filtrate (about 10 ml) by means of the TOC/DOC instrument (1.6.1.8). If the filtrate cannot be analyzed on the same day it has to be conserved acc. to 1.6.3.1. The DOC measurements (mgC/l) obtained are registered on the attached data sheet and the DOC concentrations of the culture medium and of the blanks calculated for each sampling time.

APPENDIX 2

3M Environmental Laboratory Data Sheet
-results of the Modified OECD Screening Test-

Lab Request No.: 5541 S
Date of Start of Test: 4-11-80
Analyst: Wade A. Schell
Test Material: ANTWERP FC-203, NFP Standard Material: hydroquinone, 98.5%, Lot 100147, Aldrich
Theoretical DOC of Test Material: (NOT CALCULATED)
Measured TOC[1] of Test Material: 142,000 mg TOC/kg
Inoculum: soil and non-chlorinated secondary effluent from a municipal wastewater treatment plant
Carbon Analyzer: Dohrmann DC-52A Total Organic Carbon Analyzer

A. Determination of TOC[1] of Test Material:

A stock solution of the test material was prepared by diluting 1170 mg to a final volume of 500 ml with deionized water.

conc. test material in stock solution mg/l	TOC stock solution mg/l	TOC test material mg/kg
2340	332	142,000

This result was used to calculate the necessary amount of test material to prepare a culture medium of 40 mg/l DOC[1].

B. Carbon Determinations:[2]

Culture medium	Flask no.	Conc. of test material in flask, mg/l	Calculated DOC[1] of test material in flask, mg/l	DOC Analyses	DOC - conc. after x days incubation, mg/l 0 (C_0)	7	14	21	27	28
Mineral nutrient solution with test material and with inoculum	1	282	40.0	a_1	42.4	37.0	8.1	6.2	4.1	2.7
				a_2	40.6	39.1	6.6	4.8	4.1	2.8
				a_3		36.2	6.0	7.5		
				a_4				4.2		
				a_5				4.1		
		$C_{t_1} = \frac{a_1 + a_2 \ldots a_n}{n}$			41.5	37.4	6.9	5.4	4.1	2.8
	2	282	40.0	b_1	44.0	31.8	6.5	2.8	3.2	2.9
				b_2	39.8	32.1	5.5	2.0	5.0	2.6
				b_3	41.1				4.6	
				b_4					4.2	
				b_5						
		$C_{t_2} = \frac{b_1 + b_2 \ldots b_n}{n}$			41.6	32.0	6.0	2.4	4.2	2.8
Mineral nutrient solution without test material but with inoculum	Blank 1			c_1	1.1	0.1	0.2	1.5	0.9	0.1
				c_2	1.9	-0.4	0.5	0.9	0.1	0.1
		$C_{bl_1} = \frac{c_1 + c_2}{n}$			(C_{bl_0}) 1.5	-0.2	0.4	1.2	0.5	0.1
	Blank 2			d_1	1.6	-0.1	0.1	1.6	0.4	-0.1
				d_2	1.2	0.1	0.6	1.0	0.8	-0.1
		$C_{bl_2} = \frac{d_1 + d_2}{n}$			(C_{bl_0}) 1.4	0	0.4	1.3	0.6	-0.1
				$C_{bl_{Ave}}$ =	1.4	-0.1	0.4	1.2	0.6	0

C. Evaluation of Raw Data:

Flask no.	Degradation - % DOC removal after x days incubation 7	14	21	27	28
1	6.5	83.8	89.5	91.3	93.0
2	20.1	86.1	97.0	91.0	93.0
mean	13	85	93	91	93

See attached section 1.6.3.2 for the formula used to calculate degradation.

Results of blank no.1 and no. 2 were averaged for each day x. The average values were used in the calculations.

$C_{bl_{ave}} = \frac{C_{bl_1} + C_{bl_2}}{2}$ (from above)

1. The test material is completely soluble in water. Therefore, the TOC = DOC.
2. DOC analyses performed more than twice on the same sample were run for quality assurance purposes.

3M Environmental Laboratory Data Sheet
-results of the Modified OECD Screening Test-

Lab Request No.: 5541 S
Date of Start of Test: 4-11-80
Analyst: Wade A. Schell
Test Material: ANTWERP, FC-206, FM-3824, LOT 2330, 10/79 Standard Material: hydroquinone, 98.5%, Lot 100147, Aldrich
Theoretical DOC of Test Material: (NOT CALCULATED)
Measured TOC[1] of Test Material: 93,900 mg TOC/kg
Inoculum: soil and non-chlorinated secondary effluent from a municipal wastewater treatment plant
Carbon Analyzer: Dohrmann DC-52A Total Organic Carbon Analyzer

A. Determination of TOC[1] of Test Material:

A stock solution of the test material was prepared by diluting 1,060 mg to a final volume of 500 ml with deionized water.

conc. test material in stock solution mg/l	TOC stock solution mg/l	TOC test material mg/kg
2,120	199	93,900

This result was used to calculate the necessary amount of test material to prepare a culture medium of 40 mg/l DOC[1].

B. Carbon Determinations:[2]

Culture medium	Flask no.	Conc. of test material in flask, mg/l	Calculated DOC[1] of test material in flask, mg/l	DOC Analyses	DOC - conc. after x days incubation, mg/l 0 (C_o)	7	14	21	27	28
Mineral nutrient solution with test material and with inoculum	1	426	40.0	a_1	42.3	31.0	5.3	3.6	3.4	3.0
				a_2	41.7	31.9	6.1	3.5	3.4	4.5
				a_3						3.0
				a_4						
				a_5						
		$C_{t_1} = \frac{a_1 + a_2 \ldots a_n}{n}$			42.0	31.0	5.7	3.6	3.4	3.5
	2	426	40.0	b_1	42.3	30.9	3.5	3.0	4.1	2.6
				b_2	41.3	30.7	3.3	4.1	3.2	3.6
				b_3						
				b_4						
				b_5						
		$C_{t_2} = \frac{b_1 + b_2 \ldots b_n}{n}$			41.8	30.8	3.4	3.6	3.6	3.1
Mineral nutrient solution without test material but with inoculum	Blank 1			c_1	1.1	0.1	0.2	1.5	0.9	0.1
				c_2	1.9	-0.4	0.5	0.9	0.1	0.1
		$C_{bl_1} = \frac{c_1 + c_2}{n}$			(C_{bl_0}) 1.5	-0.2	0.4	1.2	0.5	0.1
	Blank 2			d_1	1.6	-0.1	0.1	1.6	0.4	-0.1
				d_2	1.2	0.1	0.6	1.0	0.8	-0.1
		$C_{bl_2} = \frac{d_1 + d_2}{n}$			(C_{bl_0}) 1.4	0	0.4	1.3	0.6	-0.1
			$C_{bl_{Ave}} =$		1.4	-0.1	0.4	1.2	0.6	0

C. Evaluation of Raw Data:

Flask no.	Degradation - % DOC removal after x days incubation 7	14	21	27	28
1	23.3	86.9	94.1	93.1	91.4
2	23.5	92.6	94.1	92.6	92.3
mean	23	90	94	93	92

See attached section 1.6.3.2 for the formula used to calculate degradation.

Results of blank no. 1 and no. 2 were averaged for each day x. The average values were used in the calculations,

$C_{bl_{ave}} = \frac{C_{bl_1} + C_{bl_2}}{2}$ (from above)

1. The test material is completely soluble in water. Therefore, the TOC = DOC.
2. DOC analyses performed more than twice on the same sample were run for quality assurance purposes.

3M Environmental Laboratory Data Sheet
-results of the Modified OECD Screening Test-

Lab Request No.: 5541 S
Date of Start of Test: 4-11-80
Analyst: Wade A. Schell
Test Material: ANTWERP, FC-3017, LOT-2303 Standard Material: hydroquinone, 98.5%, Lot 100147, Aldrich
Theoretical DOC of Test Material: (NOT CALCULATED)
Measured TOC[1] of Test Material: 192,000 mg TOC/kg
Inoculum: soil and non-chlorinated secondary effluent from a municipal wastewater treatment plant
Carbon Analyzer: Dohrmann DC-52A Total Organic Carbon Analyzer

A. Determination of TOC[1] of Test Material:

A stock solution of the test material was prepared by diluting 1,245 mg to a final volume of 500 ml with deionized water.

conc. test material in stock solution mg/l	TOC stock solution mg/l	TOC test material mg/kg
2,490	479	192,000

This result was used to calculate the necessary amount of test material to prepare a culture medium of 40 mg/l DOC[1].

B. Carbon Determinations:[2]

Culture medium	Flask no.	Conc. of test material in flask, mg/l	Calculated DOC[1] of test material in flask, mg/l	DOC Analyses	DOC - conc. after x days incubation, mg/l 0 (C_o)	7	14	21	27	28
Mineral nutrient solution with test material and with inoculum	1	208	40.0	a_1	41.4	31.2	2.4	3.9	1.9	2.0
				a_2	40.8	30.3	2.5	2.2	3.2	1.0
				a_3					2.5	
				a_4						
				a_5						
		$C_{t_1} = \frac{a_1 + a_2 \ldots a_n}{n}$			41.1	30.8	2.4	3.0	2.5	1.5
	2	208	40.0	b_1	42.3	18.4	3.5	3.9	1.7	2.1
				b_2	41.2	19.9	3.8	1.9	2.2	2.4
				b_3						
				b_4						
				b_5						
		$C_{t_2} = \frac{b_1 + b_2 \ldots b_n}{n}$			41.8	19.2	3.6	2.9	2.0	2.2
Mineral nutrient solution without test material but with inoculum	Blank 1			c_1	1.1	0.1	0.2	1.5	0.9	0.1
				c_2	1.9	-0.4	0.5	0.9	0.1	0.1
		$C_{bl_1} = \frac{c_1 + c_2}{n}$			(C_{bl_0}) 1.5	-0.2	0.4	1.2	0.5	0.1
	Blank 2			d_1	1.6	-0.1	0.1	1.6	0.4	-0.1
				d_2	1.2	0.1	0.6	1.0	0.8	-0.1
		$C_{bl_2} = \frac{d_1 + d_2}{n}$			(C_{bl_0}) 1.4	0	0.4	1.3	0.6	-0.1
C. Evaluation of Raw Data:			$C_{bl\,ave}$ =		1.4	-0.1	0.4	1.2	0.6	0

Flask no.	Degradation - % DOC removal after x days incubation 7	14	21	27	28
1	22.2	95.0	95.5	95.2	96.2
2	52.2	92.1	95.8	96.5	94.6
mean	37	94	96	96	95

See attached section 1.6.3.2 for the formula used to calculate degradation.

Results of blank no.1 and no. 2 were averaged for each day x. The average values were used in the calculations.

$C_{bl\,ave} = \frac{C_{bl_1} + C_{bl_2}}{2}$ (from above)

1. The test material is completely soluble in water. Therefore, the TOC = DOC.
2. DOC analyses performed more than twice on the same sample were run for quality assurance purposes.

3M Environmental Laboratory Data Sheet
-results of the Modified OECD Screening Test-

Lab Request No.: 5541 S
Date of Start of Test: 4-11-80
Analyst: Wade A. Schell
Test Material: _____ Standard Material: hydroquinone, 98.5%, Lot 100147, Aldrich
Theoretical DOC of Test Material: 65.4% C (ALL CARBON ASSUMED TO BE SOLUBLE ORGANIC CARBON)
Measured TOC[1] of Test Material: _____ mg TOC/kg
Inoculum: soil and non-chlorinated secondary effluent from a municipal wastewater treatment plant
Carbon Analyzer: Dohrmann DC-52A Total Organic Carbon Analyzer

A. Determination of TOC[1] of Test Material:

A stock solution of the test material was prepared by diluting _____ mg to a final volume of _____ ml with deionized water.

conc. test material in stock solution mg/l	TOC stock solution mg/l	TOC test material mg/kg

This result was used to calculate the necessary amount of test material to prepare a culture medium of 40 mg/l DOC[1].

EVALUATION OF CALIBRATION COMPOUND - SEE STANDARD MATERIAL ABOVE.

B. Carbon Determinations:[2]

Culture medium	Flask no.	Conc. of test material in flask, mg/l	Calculated DOC[1] of test material in flask, mg/l	DOC Analyses	DOC - conc. after x days incubation, mg/l 0 (C_o)	7	14	21	27	28
Mineral nutrient solution with test material and with inoculum	1	30.6	20.0	a_1	17.1	2.3	2.0	2.4	1.3	1.7
				a_2	16.3	1.7	1.2	2.3	1.9	1.0
				a_3	18.4			3.3		
				a_4	16.2					
				a_5	18.6					
		$C_{t_1} = \frac{a_1 + a_2 \ldots a_n}{n}$			17.3	2.0	1.6	2.7	1.6	1.4
	2	30.6	20.0	b_1	17.8	2.6	1.5	0.5	1.5	0.8
				b_2	16.2	0.5	2.4	1.2	1.4	0.8
				b_3		0.6	1.6			
				b_4						
				b_5						
		$C_{t_2} = \frac{b_1 + b_2 \ldots b_n}{n}$			17.0	1.2	1.8	0.8	1.4	0.8
Mineral nutrient solution without test material but with inoculum	Blank 1			c_1	1.1	0.1	0.2	1.5	0.9	0.1
				c_2	1.9	-0.4	0.5	0.4	0.1	0.1
		$C_{bl_1} = \frac{c_1 + c_2}{n}$			(C_{bl_0}) 1.5	-0.2	0.4	1.2	0.5	0.1
	Blank 2			d_1	1.6	-0.1	0.1	1.6	0.4	-0.1
				d_2	1.2	0.1	0.6	1.0	0.8	-0.1
		$C_{bl_2} = \frac{d_1 + d_2}{n}$			(C_{bl_0}) 1.4	0	0.4	1.3	0.6	-0.1
C. Evaluation of Raw Data:			$C_{bl_{ave}} = \frac{C_{bl_1} + C_{bl_2}}{2} =$		1.4	-0.1	0.4	1.2	0.6	0

Flask no.	Degradation - % DOC removal after x days incubation 7	14	21	27	28
1	86.8	92.5	90.6	93.7	91.2
2	91.7	91.0	102.6	94.9	94.9
mean	89	92	97	94	93

See attached section 1.6.3.2 for the formula used to calculate degradation.

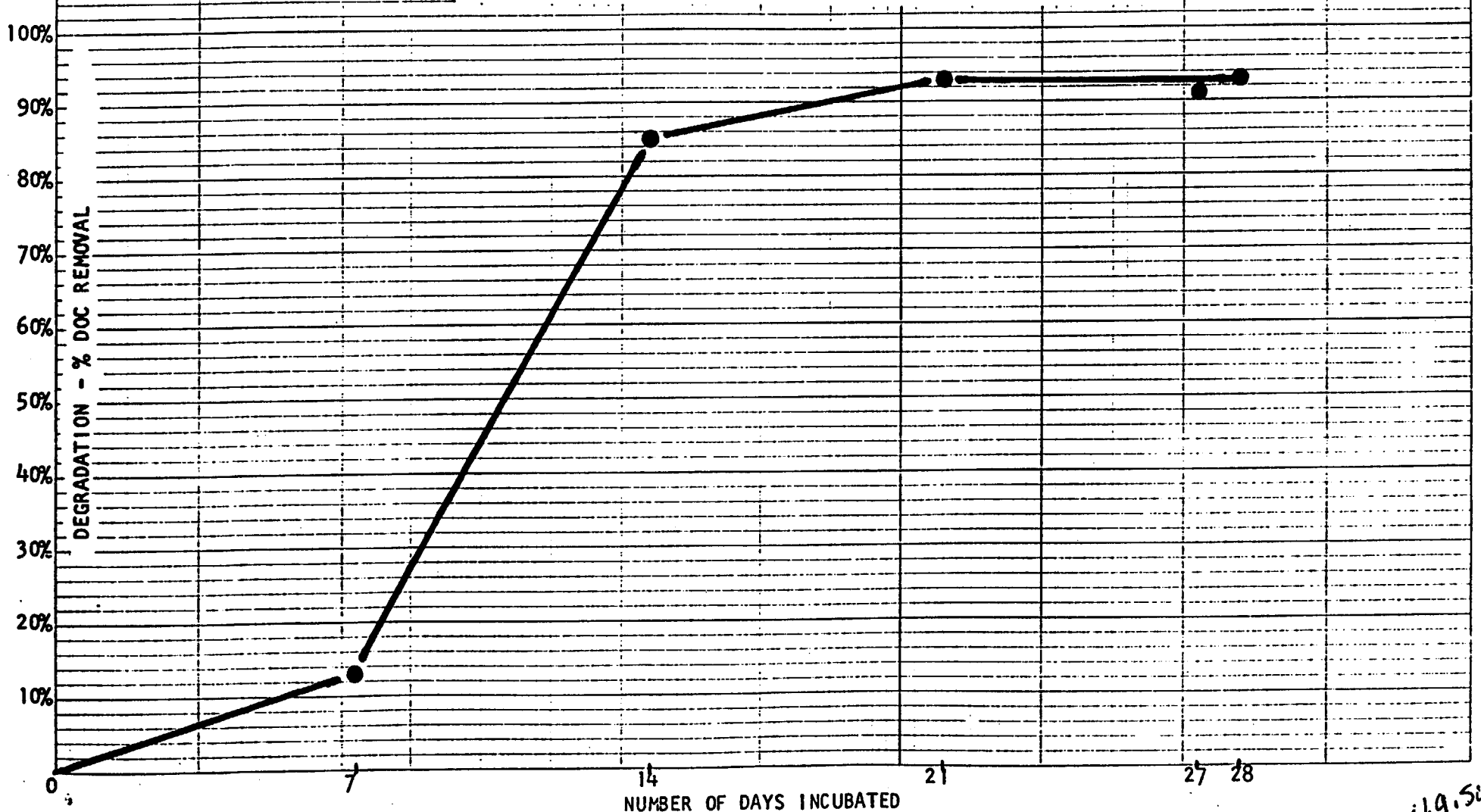
Results of blank no.1 and no. 2 were averaged for each day x. The average values were used in the calculations

$C_{bl_{ave}} = \frac{C_{bl_1} + C_{bl_2}}{2}$ (from above)

1. The test material is completely soluble in water. Therefore, the TOC = DOC.
2. DOC analyses performed more than twice on the same sample were run for quality assurance purposes.

3M Environmental Laboratory
OECD Modified Screening Test with DOC Analysis
Analyst: Wade A. Schell
Date of Start of Test: 4-11-1980
Lab Request No.: 5541 S

Test Material: ANTWERP, FC-203, NFP

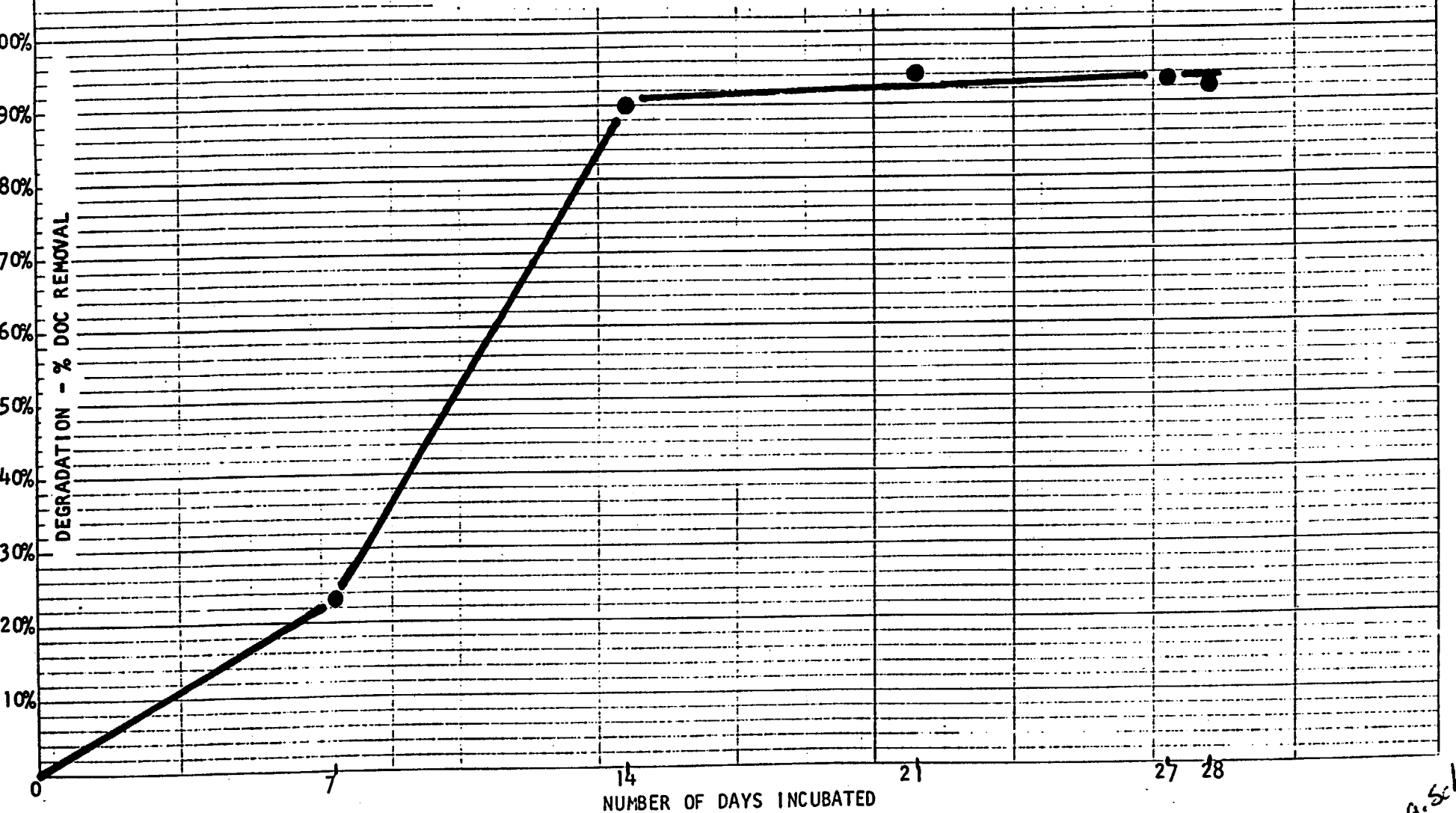


W.A. Schell
5-23-80

APPENDIX 3

3M Environmental Laboratory
OECD Modified Screening Test with DOC Analysis
Analyst: Wade A. Schell
Date of Start of Test: 4-11-1980
Lab Request No.: 5541 S

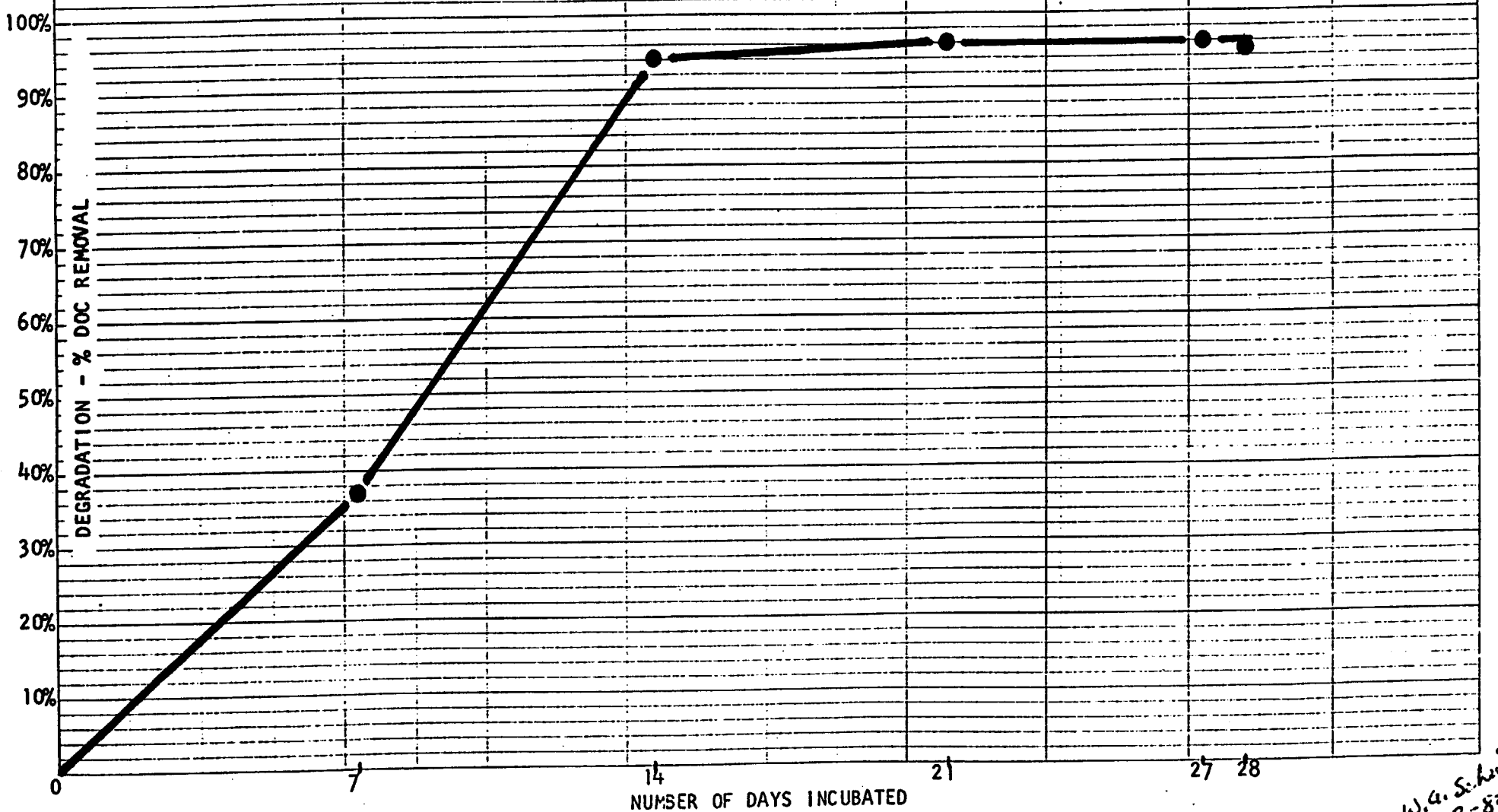
Test Material: ANTWERP, FC-206, FM-3824
LOT-2330, 10/79



W.A. Schell
5-23-80

3M Environmental Laboratory
OECD Modified Screening Test with DOC Analysis
Analyst: Wade A. Schell
Date of Start of Test: 4-11-1980
Lab Request No.: 5541 S

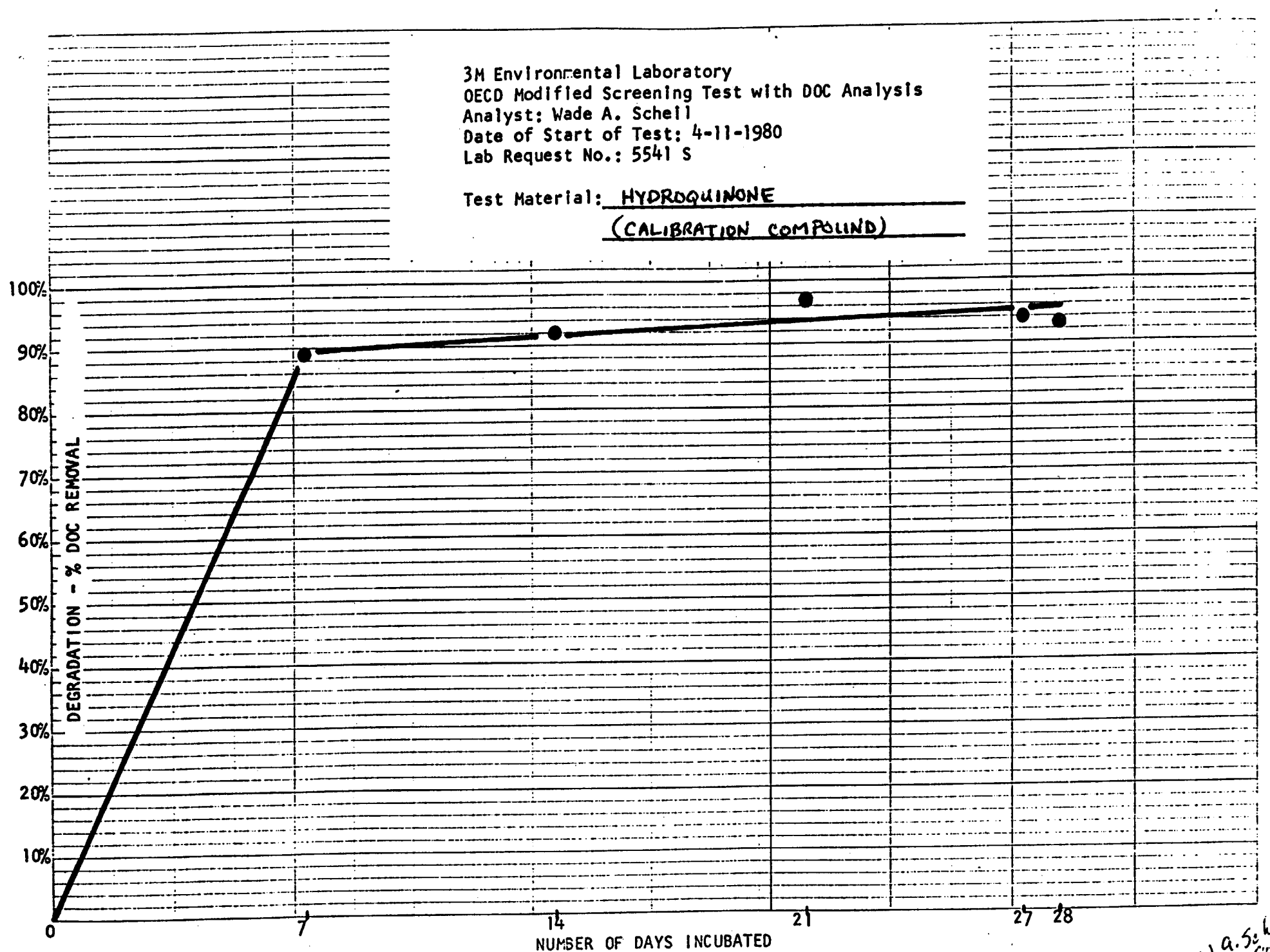
Test Material: ANTWERP, FC-3017, LOT-2303



W.A. Schell
5-23-80

3M Environmental Laboratory
OECD Modified Screening Test with DOC Analysis
Analyst: Wade A. Schell
Date of Start of Test: 4-11-1980
Lab Request No.: 5541 S

Test Material: HYDROQUINONE
(CALIBRATION COMPOUND)



W.A.S. 5-23-80

LR # 55415

September 19., 1979

OECD Chemicals Testing Programme

Test Guideline for the Modified
OECD Screening Test with DOC Analysis

0. The test procedure constitutes a modification of the OECD Screening Test (OECD Environment Directorate, Proposed Method for the Determination of the Biodegradability of Surfactants Used in Synthetic Detergents, Paris 1976, and council directive of Nov. 22, 1973, on the approximation of the laws of the member states relating to methods of testing the biodegradability of anionic surfactants (73/405/EEC), Official Journal of the European Communities No. 4 347/53 of Dec. 17, 1973) for the application of the dissolved organic carbon (DOC) analysis.

1. Method

1.1 Introduction: Purpose, scope, relevance, and application of test and explanation of limits.

The purpose of the method is the measurement of the ultimate biodegradability of water soluble, non - volatile organic compounds in an aerobic, aqueous medium at a starting test concentration corresponding to 5 - 40 mg DOC/l (In order to avoid inhibitory effects it is in the investigator's own interest

3M Environmental Laboratory Data Sheet
-results of the Modified OECD Screening Test-

Lab Request No.: 5541 S
Date of Start of Test: 4-11-80
Analyst: Wade A. Schell
Test Material: ANTWERP FC-203, NFP Standard Material: hydroquinone, 98.5%, Lot 100147, Aldrich
Theoretical DOC of Test Material: (NOT CALCULATED) ~~0.65 g/g~~
Measured TOC[1] of Test Material: 142,000 mg TOC/kg
Inoculum: soil and non-chlorinated secondary effluent from a municipal wastewater treatment plant
Carbon Analyzer: Dohrmann DC-52A Total Organic Carbon Analyzer

A. Determination of TOC[1] of Test Material:

A stock solution of the test material was prepared by diluting 1170 mg to a final volume of 500 ml with deionized water.

conc. test material in stock solution mg/l	TOC stock solution mg/l	TOC test material mg/kg
2340	332	142,000

This result was used to calculate the necessary amount of test material to prepare a culture medium of 40 mg/l DOC[1].

B. Carbon Determinations:[2]

Culture medium	Flask no.	Conc. of test material in flask, mg/l	Calculated DOC[1] of test material in flask, mg/l	DOC Analyses	DOC - conc. after x days incubation, mg/l 0 (C_o)	7	14	21	27	28
Mineral nutrient solution with test material and with inoculum	1	282	40.0	a_1	42.4	37.0	8.1	6.2	4.1	2.7
				a_2	40.6	39.1	6.6	4.8	4.1	2.8
				a_3		36.2	6.0	7.5		
				a_4				4.2		
				a_5				4.1		
		$C_{t_1} = \frac{a_1 + a_2 \ldots a_n}{n}$			41.5	37.4	6.9	5.4	4.1	2.8
	2	282	40.0	b_1	44.0	31.8	6.5	2.8	3.2	2.9
				b_2	39.8	32.1	5.5	2.0	5.0	2.6
				b_3	41.1				4.6	
				b_4					4.2	
				b_5						
		$C_{t_2} = \frac{b_1 + b_2 \ldots b_n}{n}$			41.6	32.0	6.0	2.4	4.2	2.8
Mineral nutrient solution without test material but with inoculum	Blank 1			c_1	1.1	0.1	0.2	1.5	0.9	0.1
				c_2	1.9	-0.4	0.5	0.9	0.1	0.1
		$C_{bl_1} = \frac{c_1 + c_2}{n}$			(C_{bl_0}) 1.5	-0.2	0.4	1.2	0.5	0.1
	Blank 2			d_1	1.6	-0.1	0.1	1.6	0.4	-0.1
				d_2	1.2	0.1	0.6	1.0	0.8	-0.
		$C_{bl_2} = \frac{d_1 + d_2}{n}$			(C_{bl_0}) 1.4	0	0.4	1.3	0.6	-0.
				$C_{bl_{Ave}}$ =	1.4	-0.1	0.4	1.2	0.6	0

C. Evaluation of Raw Data:

Flask no.	Degradation - % DOC removal after x days incubation 7	14	21	27	28
1	6.5	83.8	89.5	91.3	93.0
2	20.1	86.1	97.0	91.0	93.0
mean	13	85	93	91	93

See attached section 1.6.3.2 for the formula used to calculate degradation.

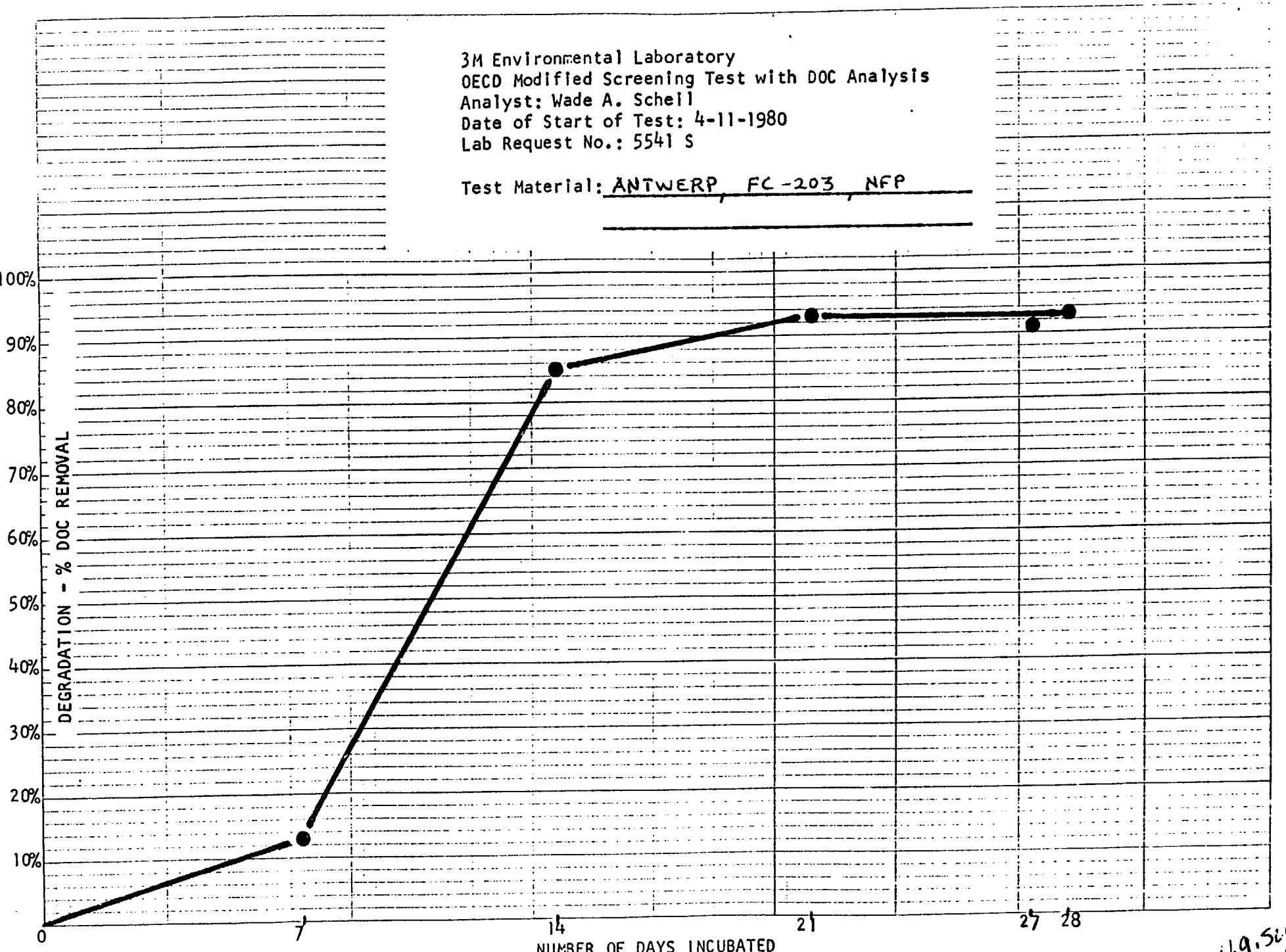
Results of blank no.1 and no. 2 were averaged for each day x. The average values were used in the calculations.

$C_{bl_{ave}} = \frac{C_{bl_1} + C_{bl_2}}{2}$ (from above)

1. The test material is completely soluble in water. Therefore, the TOC = DOC.
2. DOC analyses performed more then twice on the same sample were run for quality assurance purposes.

3M Environmental Laboratory
OECD Modified Screening Test with DOC Analysis
Analyst: Wade A. Schell
Date of Start of Test: 4-11-1980
Lab Request No.: 5541 S

Test Material: ANTWERP, FC-203, NFP



W.A. Schell
5-23-80

3M Environmental Laboratory Data Sheet
-results of the Modified OECD Screening Test-

Lab Request No.: 5541 S
Date of Start of Test: 4-11-80
Analyst: Wade A. Schell
Test Material: ANTWERP, FC-3017, LOT-2303 Standard Material: hydroquinone, 98.5%, Lot 100147, Aldrich
Theoretical DOC of Test Material: (NOT CALCULATED)
Measured TOC[1] of Test Material: 192,000 mg TOC/kg
Inoculum: soil and non-chlorinated secondary effluent from a municipal wastewater treatment plant
Carbon Analyzer: Dohrmann DC-52A Total Organic Carbon Analyzer

A. Determination of TOC[1] of Test Material:

A stock solution of the test material was prepared by diluting 1,245 mg to a final volume of 500 ml with deionized water.

conc. test material in stock solution mg/l	TOC stock solution mg/l	TOC test material mg/kg
2,490	479	192,000

This result was used to calculate the necessary amount of test material to prepare a culture medium of 40 mg/l DOC[1].

B. Carbon Determinations:[2]

Culture medium	Flask no.	Conc. of test material in flask, mg/l	Calculated DOC[1] of test material in flask, mg/l	DOC Analyses	DOC - conc. after x days incubation, mg/l: 0 (C_o)	7	14	21	27	28
Mineral nutrient solution with test material and with inoculum	1	208	40.0	a_1	41.4	31.2	2.4	3.9	1.9	2.0
				a_2	40.8	30.3	2.5	2.2	3.2	1.0
				a_3					2.5	
				a_4						
				a_5						
		$C_{t_1} = \frac{a_1 + a_2 \cdots a_n}{n}$			41.1	30.8	2.4	3.0	2.5	1.5
	2	208	40.0	b_1	42.3	18.4	3.5	3.9	1.7	2.1
				b_2	41.2	19.9	3.8	1.8	2.2	2.4
				b_3						
				b_4						
				b_5						
		$C_{t_2} = \frac{b_1 + b_2 \cdots b_n}{n}$			41.8	19.2	3.6	2.9	2.0	2.2
Mineral nutrient solution without test material but with inoculum	Blank 1			c_1	1.1	0.1	0.2	1.5	0.9	0.1
				c_2	1.9	-0.4	0.5	0.9	0.1	0.1
		$C_{bl_1} = \frac{c_1 + c_2}{n}$			(C_{bl_0}) 1.5	-0.2	0.4	1.2	0.5	0.1
	Blank 2			d_1	1.6	-0.1	0.1	1.6	0.4	-0.1
				d_2	1.2	0.1	0.6	1.0	0.8	-0.1
		$C_{bl_2} = \frac{d_1 + d_2}{n}$			(C_{bl_0}) 1.4	0	0.4	1.3	0.6	-0.1
			$C_{bl\,ave} =$		1.4	-0.1	0.4	1.2	0.6	0

C. Evaluation of Raw Data:

Flask no.	Degradation - % DOC removal after x days incubation: 7	14	21	27	28
1	22.2	95.0	95.5	95.2	96.2
2	52.2	92.1	95.8	96.5	94.6
mean	37	94	96	96	95

See attached section 1.6.3.2 for the formula used to calculate degradation.

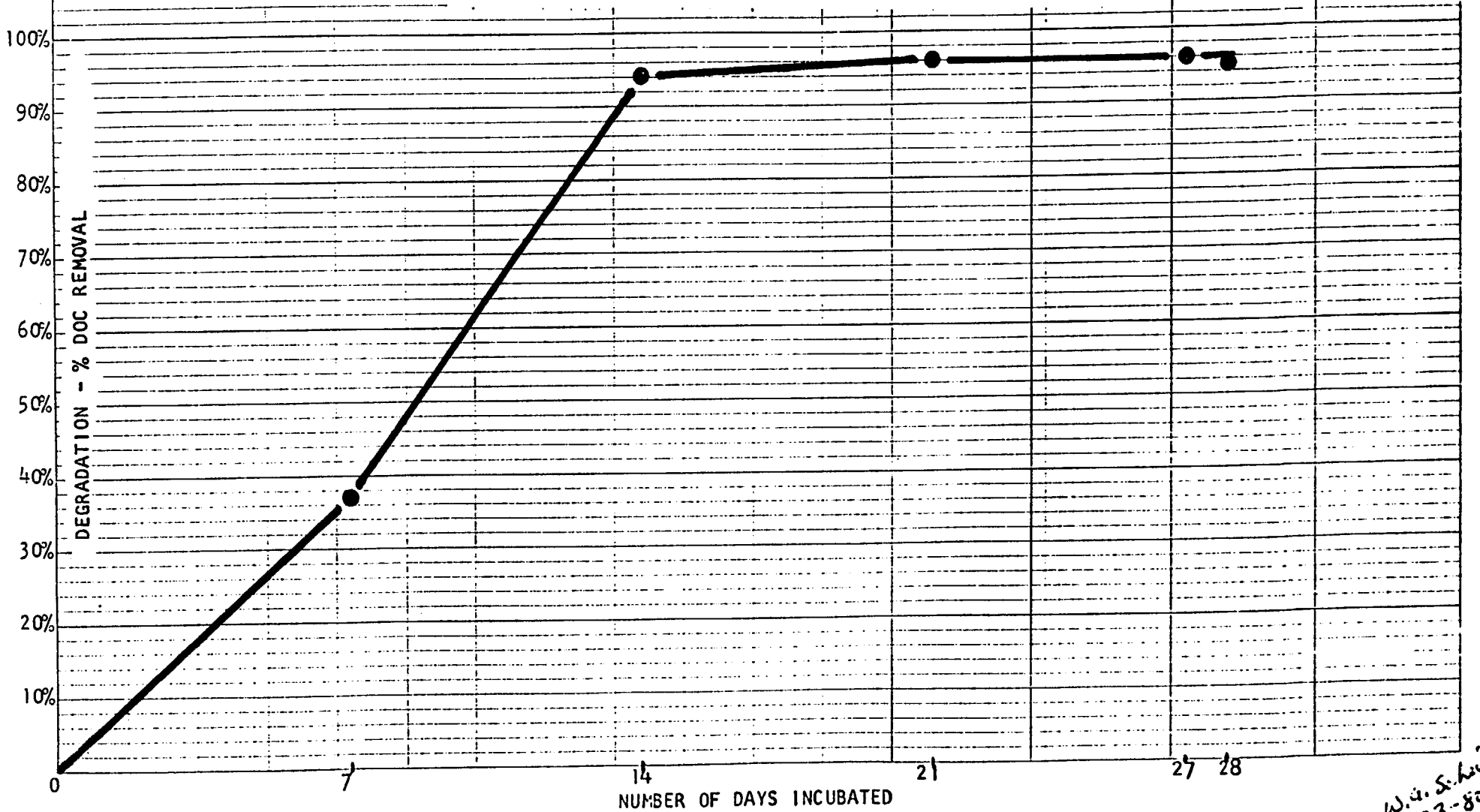
Results of blank no.1 and no. 2 were averaged for each day x. The average values were used in the calculations.

$C_{bl_{ave}} = \frac{C_{bl_1} + C_{bl_2}}{2}$ (from above)

1. The test material is completely soluble in water. Therefore, the TOC = DOC.
2. DOC analyses performed more than twice on the same sample were run for quality assurance purposes.

3M Environmental Laboratory
OECD Modified Screening Test with DOC Analysis
Analyst: Wade A. Scheil
Date of Start of Test: 4-11-1980
Lab Request No.: 5541 S

Test Material: ANTWERP, FC-3017, LOT-2303



W.A. Scheil
5-23-80

3M Environmental Laboratory Data Sheet
-results of the Modified OECD Screening Test-

Lab Request No.: 5541 S
Date of Start of Test: 4-11-80
Analyst: Wade A. Schell
Test Material: ANTWERP, FC-206, FM-3824, LOT 2330, 10/79 Standard Material: hydroquinone, 98.5%, Lot 100147, Aldrich
Theoretical DOC of Test Material: (NOT CALCULATED)
Measured TOC[1] of Test Material: 93,900 mg TOC/kg
Inoculum: soil and non-chlorinated secondary effluent from a municipal wastewater treatment plant
Carbon Analyzer: Dohrmann DC-52A Total Organic Carbon Analyzer

A. Determination of TOC[1] of Test Material:

A stock solution of the test material was prepared by diluting 1,060 mg to a final volume of 500 ml with deionized water.

conc. test material in stock solution mg/l	TOC stock solution mg/l	TOC test material mg/kg
2,120	199	93,900

This result was used to calculate the necessary amount of test material to prepare a culture medium of 40 mg/l DOC[1].

B. Carbon Determinations:[2]

Culture medium	Flask no.	Conc. of test material in flask, mg/l	Calculated DOC[1] of test material in flask, mg/l	DOC Analyses	DOC - conc. after x days incubation, mg/l 0 (C_o)	7	14	21	27	28
Mineral nutrient solution with test material and with inoculum	1	426	40.0	a_1	42.3	31.0	5.3	3.6	3.4	3.0
				a_2	41.7	31.0	6.1	3.5	3.4	4.5
				a_3						3.0
				a_4						
				a_5						
		$C_{t_1} = \frac{a_1 + a_2 \ldots a_n}{n}$			42.0	31.0	5.7	3.6	3.4	3.5
	2	426	40.0	b_1	42.3	30.9	3.5	3.0	4.1	2.6
				b_2	41.3	30.7	3.3	4.1	3.2	3.6
				b_3						
				b_4						
				b_5						
		$C_{t_2} = \frac{b_1 + b_2 \ldots b_n}{n}$			41.8	30.8	3.4	3.6	3.6	3.1
Mineral nutrient solution without test material but with inoculum	Blank 1			c_1	1.1	0.1	0.2	1.5	0.9	0.1
				c_2	1.9	-0.4	0.5	0.9	0.1	0.1
		$C_{bl_1} = \frac{c_1 + c_2}{n}$			(C_{bl_0}) 1.5	-0.2	0.4	1.2	0.5	0.1
	Blank 2			d_1	1.6	-0.1	0.1	1.6	0.4	-0.1
				d_2	1.2	0.1	0.6	1.0	0.8	-0.1
		$C_{bl_2} = \frac{d_1 + d_2}{n}$			(C_{bl_0}) 1.4	0	0.4	1.3	0.6	-0.1
			$C_{bl_{Ave}} =$		1.4	-0.1	0.4	1.2	0.6	0

C. Evaluation of Raw Data:

Flask no.	Degradation - % DOC removal after x days incubation 7	14	21	27	28
1	23.3	86.9	94.1	93.1	91.4
2	23.5	92.6	94.1	92.6	92.3
mean	23	90	94	93	92

See attached section 1.6.3.2 for the formula used to calculate degradation.

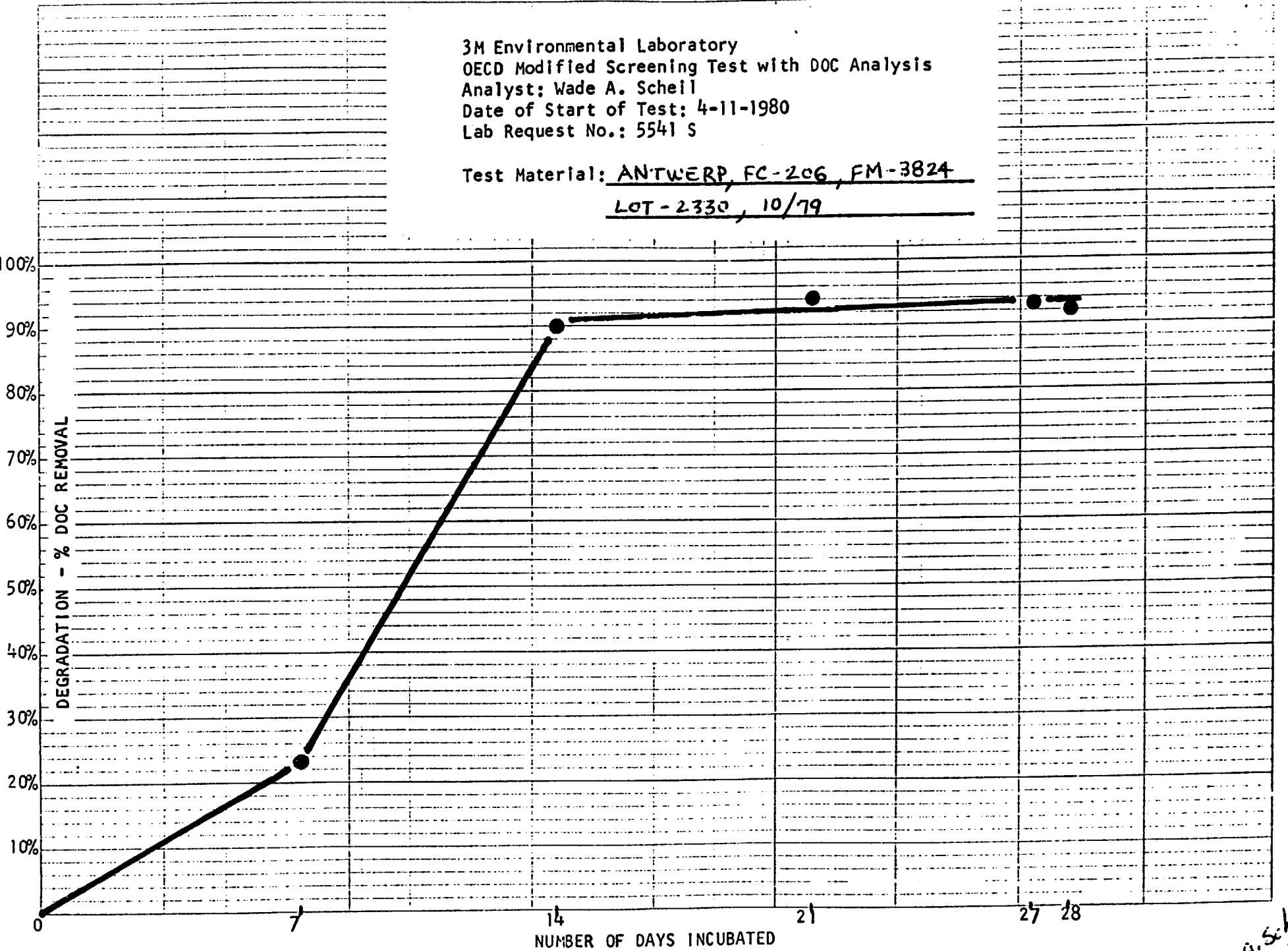
Results of blank no.1 and no. 2 were averaged for each day x. The average values were used in the calculations.

$C_{bl_{ave}} = \frac{C_{bl_1} + C_{bl_2}}{2}$ (from above)

1. The test material is completely soluble in water. Therefore, the TOC = DOC.
2. DOC analyses performed more than twice on the same sample were run for quality assurance purposes.

3M Environmental Laboratory
OECD Modified Screening Test with DOC Analysis
Analyst: Wade A. Scheil
Date of Start of Test: 4-11-1980
Lab Request No.: 5541 S

Test Material: ANTWERP, FC-206, FM-3824
LOT-2330, 10/79



W.A. Scheil
5-23-80

3M Environmental Laboratory Data Sheet
-results of the Modified OECD Screening Test-

Lab Request No.: 5541 S
Date of Start of Test: 4-11-80
Analyst: Wade A. Schell
Test Material: _____ Standard Material: hydroquinone, 98.5%, Lot 100147, Aldrich
Theoretical DOC of Test Material: 65.4% C (ALL CARBON ASSUMED TO BE SOLUBLE ORGANIC CARBON)
Measured TOC[1] of Test Material: _____ mg TOC/kg
Inoculum: soil and non-chlorinated secondery effluent from a municipal wastewater treatment plant
Carbon Analyzer: Dohrmann DC-52A Total Organic Carbon Analyzer

A. Determination of TOC[1] of Test Material:

A stock solution of the test material was prepared by diluting _____mg to a final volume of _____ml with deionized water.

conc. test material in stock solution mg/l	TOC stock solution mg/l	TOC test material mg/kg

This result was used to calculate the necessary amount of test material to prepare a culture medium of 40 mg/l DOC[1].

EVALUATION OF CALIBRATION COMPOUND - SEE STANDARD MATERIAL ABOVE.

B. Carbon Determinations:[2]

Culture medium	Flask no.	Conc. of test material in flask, mg/l	Calculated DOC[1] of test material in flask, mg/l	DOC Analyses	DOC - conc. after x days incubation, mg/l 0 (C_o)	7	14	21	27	28
Mineral nutrient solution with test material and with inoculum	1	30.6	20.0	a_1	17.1	2.3	2.0	2.4	1.3	1.7
				a_2	16.3	1.7	1.2	2.3	1.9	1.0
				a_3	18.4			3.3		
				a_4	16.2					
				a_5	18.6					
		$C_{t_1} = \frac{a_1 + a_2 \ldots a_n}{n}$			17.3	2.0	1.6	2.7	1.6	1.4
	2	30.6	20.0	b_1	17.8	2.6	1.5	0.5	1.5	0.8
				b_2	16.2	0.5	2.4	1.2	1.4	0.8
				b_3		0.6	1.6			
				b_4						
				b_5						
		$C_{t_2} = \frac{b_1 + b_2 \ldots b_n}{n}$			17.0	1.2	1.8	0.8	1.4	0.8
Mineral nutrient solution without test material but with inoculum	Blank 1			c_1	1.1	0.1	0.2	1.5	0.9	0.1
				c_2	1.9	-0.4	0.5	0.9	0.1	0.1
		$C_{bl_1} = \frac{c_1 + c_2}{n}$			(C_{bl_0}) 1.5	-0.2	0.4	1.2	0.5	0.1
	Blank 2			d_1	1.6	-0.1	0.1	1.6	0.4	-0.1
				d_2	1.2	0.1	0.6	1.0	0.8	-0.1
		$C_{bl_2} = \frac{d_1 + d_2}{n}$			(C_{bl_0}) 1.4	0	0.4	1.3	0.6	-0.1
			$C_{bl_{ave}} = \frac{C_{bl_1} + C_{bl_2}}{2} =$		1.4	-0.1	0.4	1.2	0.6	0

C. Evaluation of Raw Data:

Flask no.	Degradation - % DOC removal after x days incubation 7	14	21	27	28
1	86.8	92.5	90.6	93.7	91.2
2	91.7	91.0	102.6	94.9	94.9
mean	89	92	97	94	93

See attached section 1.6.3.2 for the formula used to calculate degradation.

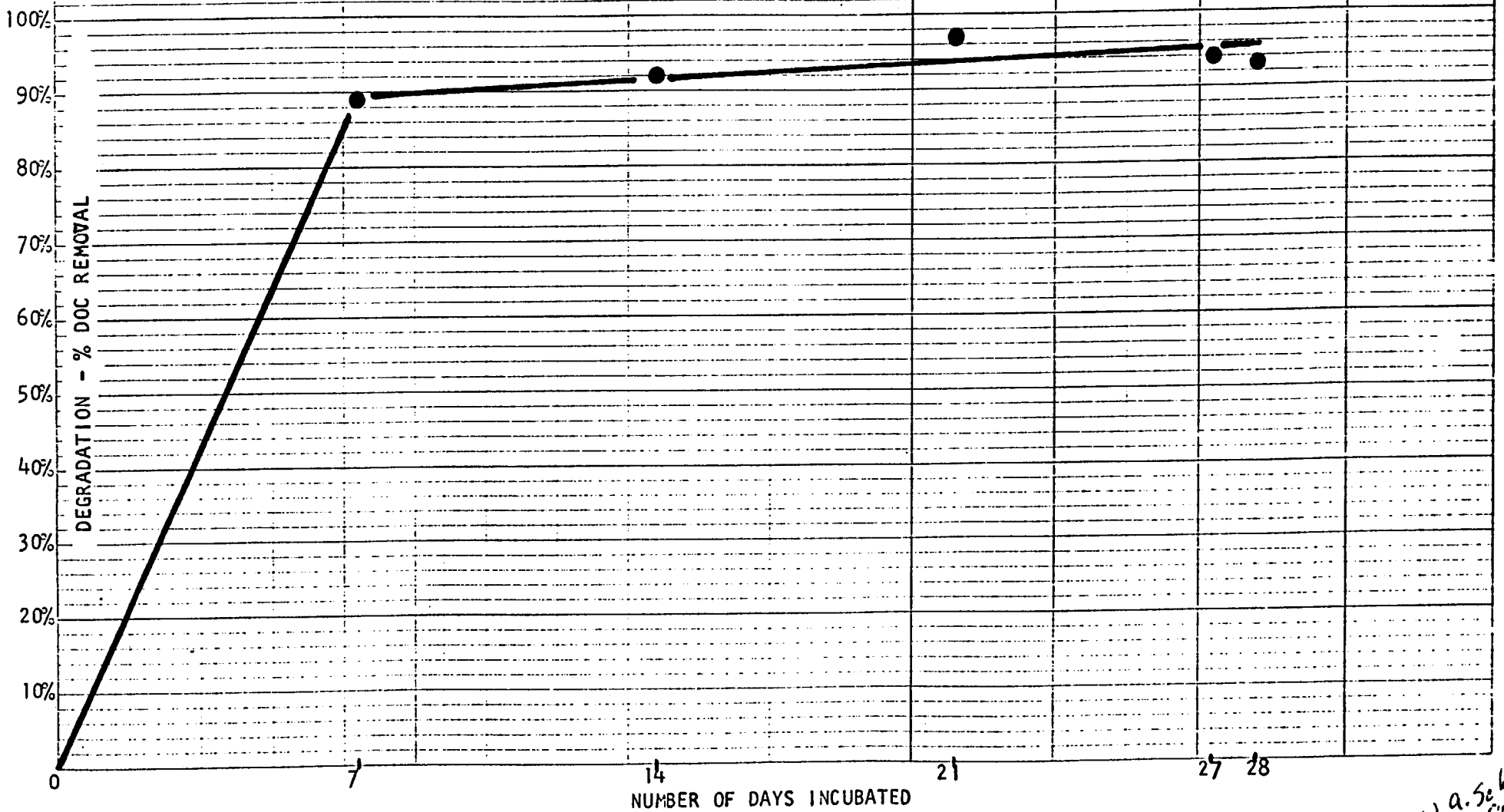
Results of blank no.1 and no. 2 were averaged for each day x. The average values were used in the calculations

$C_{bl_{ave}} = \frac{C_{bl_1} + C_{bl_2}}{2}$ (from above)

1. The test material is completely soluble in water. Therefore, the TOC = DOC.
2. DOC analyses performed more than twice on the same sample were run for quality assurance purposes.

3M Environmental Laboratory
OECD Modified Screening Test with DOC Analysis
Analyst: Wade A. Scheil
Date of Start of Test: 4-11-1980
Lab Request No.: 5541 S

Test Material: HYDROQUINONE
(CALIBRATION COMPOUND)



W. A. Scheil
5-23-80

LR5541 S.

FORM 10620-8-PWO

File: Page No. 1 of 5

ENVIRONMENTAL ENGR. LAB – WORK SHEET

SAMPLE DESCRIPTION

Date: 4-08-80
Analyst: Wade A. S. Cmd
Hrs.:

Notes – OECD Screening Test (Modified)

1. Nutrient solution preparation:

✓ a) Anhydrous Na_2HPO_4 was used. FW = 141.96
$2H_2O = 36.0$, $\frac{142}{178} = 0.798$ correction ratio.

✓ b) $CaCl_2 \cdot 2H_2O$ was used. FW = 147.03
correction ratio: $\frac{147}{111} = 1.32$

✓ c) Yeast extract was used instead of solution 1.6.1.2 (F)

2. 0.10g $HgCl_2$ per 250 ml test solution was used for sterile controls.

✓ 3. Reaction vessels were filled with 250 ml of test solution, and they were inoculated with 0.14 ml inoculum.

✓ 4. Inoculum: Pigs eye mixed liquor – decanted, filtered through Whatmann® 54 paper filter and kept aerated until use. Soil extract (1.6.2.2), – Whatmann® 54 a 50-50 composite of the two (2) above sources was used for testing.
NOTE: Soil source – E.A. Poinier's garden.
1.6.2.4. Composite inoculum, 50-50 of 1.6.2.1 and 1.6.2.2

5. Before filtering of test solutions on day-0, test conc 4-3 formed a white precipitate.

Reviewed by:

LR # 5541-5

FORM 10620-8-PWO

File: Page No. 2 of 5

ENVIRONMENTAL ENGR. LAB – WORK SHEET

SAMPLE DESCRIPTION

Date: 4-14-80
Analyst: W. A. Schiel
Hrs.:

Notes – Mod. OECD Screening test.

6. On day-3 (4-14-80) considerable foaming of test solutions nos. 1-1, 1-2, 3-1 and 3-2.
7. On day-7 (4-18-80) foaming had somewhat subsided in test solutions nos. 1-2 and 3-2.
8. On day-10 (4-21-80) foaming had almost entirely subsided on all test solutions listed in note no. 6 above.
9. On day-14 (4-25-80) foaming had resumed in test solution noted in no. 6 above.
10. On day-21 (5-02-80) foaming on test solutions noted in no. 6 above persisted.
The solution in reactor vessel 4-3 was slightly brownish color. Also the precipitate in the vessel appeared slightly brownish.
11. On day-27 (5-08-80) and day-28 (5-09-80) the solutions in glass nos. 1-1, 1-2, 2-2, 3-1, and 3-2 were foaming.
~~12. Procedure of filtration and preservation techniques:~~

Reviewed by:

FORM 10620-8-PWO

File: LR Page No. 5541 S

Page 3 of 5

ENVIRONMENTAL ENGR. LAB – WORK SHEET

SAMPLE DESCRIPTION

Date: 5-23-80
Analyst: W. G. Scheel
Hrs.:

Note on Mod. OECD Screening Test

12. The hydroquinone (calibration compound) used was 98.5%. Better quality reagent could not be obtained from common suppliers. 4 Chemical suppliers were tried
13. Filters used: 0.20 um Gelman® membrane filters, part no. 64814
14. Chlorine-free well water (Env. Lab bioassay room) was used to extract soil for inoculum preparation. The extract was filtered through Whatman® #54 paper filter paper.
15. Cultures were incubated in the dark.
16. The incubation temperature on the 6th day exceeded the maximum of 25°C by ~0.5°C for an ~3 HR period – see temperature strip chart for 4-17-80.
17. Preparation of Trace element solution:
    Fe-chelate – 30 mg $FeCl_3$ and 70 mg EDTA were used
18. All glassware was cleaned with Chromerge® (chromic-acid cleaning solution) and rinsed at least six (6) times with D.I. H_2O.
19. Inoculum – the secondary effluent sample was obtained by taking a sample out of a secondary aeration basin and allowing the sludge to settle and then decanting. – see note no. 4 above.
20. Clean porous foam plugs were used to stopper the aeration flasks.
21. Sample withdrawal – at the various test days the volume of the culture medium was adjusted to compensate for evaporate

Reviewed by:

cont. page 4

e-PWO

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page 4 of 5

ENVIRONMENTAL ENGR. LAB – WORK SHEET

SAMPLE DESCRIPTION

Date: 5-23-80
Analyst: W A S. Lum
Hrs.:

continued from page 3

21. cont. –
losses by pouring the culture medium into a clean 250 ml volumetric flask, adding D.I. H_2O (1.6.1.1.) and pouring it back into the original reactor vessel.
25 ml of sample was withdrawn for each sampling period by first pouring 15ml into the filter holder and discarding this filtrate, and then pouring 10 ml into the filter holder and collecting the filtrate in a glass vial.

22. Sample preservation – immediately upon filtering the sample, 1 drop of conc. HCl was added to the 10 ml of filtered sample, bring the pH of the sample to pH < 2. The vials were capped with foil lined caps. and refrigerated until ~~DOC~~ analyzed for DOC.
Since acidification with HCl is a necessary step in the DOC analyses protocal it was decided by EAR and myself to acidify immediately. This same method of sample preservation is recommended for preservation of ~~used to preserve~~ the potassium hydrogen phthalate solution used to calibrate the TOC analyzer.(1) The standard is stable over extended periods of time.(1) $HgCl_2$ was not used.
LRS 5-28-80

Notes:
(1) Dohrmann DC-52A Operating Manual, 4th Ed, 1978, pg. 3-1.

Reviewed by:

LR # 5541 S

FORM 10620-6-PWO

File: Page No. 5 of 5

ENVIRONMENTAL ENGR. LAB – WORK SHEET

SAMPLE DESCRIPTION

Date: 5-23-80
Analyst: W. A. Schiff
Hrs.:

Notes - Mod. OECD Screening Test

22. A replicate reaction vessel of each of the analyzed culture mediums was prepared and $HgCl_2$ was added. These vessels served as sterile controls to ensure that volitalization of the test materials did not occur over the 28 day test period. The sterile controls were handled identically to the viable reaction vessels. Samples were withdrawn from the sterile controls as in note no. 21 above. The DOC of the sterile controls was measured for the day-0 and day-28 samples.

Results of Sterile Controls:

	DOC - mg/l		% DOC Remaining after 28 days
	DAY-0	DAY-28	
C-3 (Blank)	1.5, 1.2 } 1.4	2.1, 1.7 } 1.9	
1-3 (FC-203)	40.3, 45.1 } 42.7	38.4, 40.8 } 39.6	$\frac{(39.6-1.9)}{(42.7-1.4)} \times 100 = 91.3\%$
2-3 (FC-3017)	40.3, 41.6 } 41.0	36.7, 36.9 } 36.8	$\frac{(36.8-1.9)}{(41.0-1.4)} \times 100 = 88.1\%$
3-3 (FC-206)	40.8, 42.0 } 41.4	39.1, 39.6, 38.1 } 39.6	$\frac{(39.6-1.9)}{(41.4-1.4)} \times 100 = 94.2\%$
4-3 (Hydroquinone)	17.7, 15.0, 16.8 } 16.5	13.0, 14.2, 13.7 } 13.6	$\frac{(13.6-1.9)}{(16.5-1.4)} = 77.5\%$

$$\%\text{DOC Remaining} = \frac{(\overline{DOC} \text{ at day 28} - \overline{DOC} \text{ of Blank at day 28})}{(\overline{DOC} \text{ at day 0} - \overline{DOC} \text{ of Blank at day 0})} \times 100$$

Reviewed by:

FORM 10620-8-PWO

File: CR 5541 S
Page No. Page 1 of 1

ENVIRONMENTAL ENGR. LAB – WORK SHEET

SAMPLE DESCRIPTION

Date: 4-10-80
Analyst: W.G. Schenk
Hrs.:

Preparation of Test Solutions.

Scope: So much of each test material is diluted to 1.00 l so that the final TOC concentration is 40 mg/l.

So much hydroquinone is diluted to 1.00 l so that the TOC concentration is 20 mg/l TOC

Procedure: Weight out calculated amount and place in 1.00 l volumetric flask containing ~500 ml of nutrient solution (1.6.1.2). Dilute to 1.00 l mark with additional nutrient solution (1.6.1.2). Mix throughly and place 250 ml in each of three (3) 500 ml Erlenmeyer flasks. Discard remaining solution.

Calculated Amounts:

Sample #1, Antwerp FC-203, NFP

TOC: 142,000 mg/kg sample

$$?\,g\ sample = \frac{40\ mg\ TOC}{1} \times \frac{1.00\ Kg\ sample}{142{,}000\ mg\ TOC} \times \frac{1000\ g}{1.00\ Kg} = 0.282\ g\ FC\text{-}203$$

Sample #2, Antwerp FC-3017, LOT-2303 (192,000 mg TOC/kg sample)

$$?\,g\ sample = 40\ mg\ TOC \times \frac{1.00\ Kg\ sample}{192{,}000\ mg\ TOC} \times \frac{1000\ g}{1.00\ Kg} = 0.208\ g\ FC\text{-}3017$$

Sample # 3, Antwerp FC-206 (FM-3834) LOT 2330, 10/79

(93,900 mg TOC/kg sample)

$$?\,g\ sample = 40\ mg\ TOC \times \frac{1.00\ kg\ sample}{93{,}900\ mg\ TOC} \times \frac{1000\ g}{1.00\ kg} = 0.426\ g\ FC\text{-}206$$

Hydroquinone: 65.4% C → assume C = TOC

Reviewed by:

$$?\,g\ hydroquinone = 20\ mg\ TOC \times \frac{1.00\ g\ hydro.}{.6654\ g\ TOC} \times \frac{1.00\ g}{1000\ mg} = 0.0306\ g = 30.6\ mg$$

FILE
LR# 55415

Test Design

	CONTROL (BLANK)	CALIBRATION COMPOUND	SAMPLE #1	SAMPLE #2	SAMPLE #3
Duplicate	C-1 C-2	4-1 4-2	1-1 1-2	2-1 2-2	3-1 3-2
Sterile ($HgCl_2$)	C-3	4-3	1-3	2-3	3-3

DOC analyses must be run in duplicate.

Test Days: 0 ; ~7 , ~14 , ~21 , 27 , 28 must do underlined

# of DOC analyses: 6 test periods X 15 samples = 90 samples
90 samples analyzed in duplicate = 180 analyses

Seed:

Incubator Temp (20-25°C)(68-77°F)
Keep towards higher end of range.

Clean glassware

Prepare filters.

TOC standard

yeast Extract solution 15 mg/100 ml

FORM 10620-8-PWO

File: LR Page No. 5541 S

ENVIRONMENTAL ENGR. LAB – WORK SHEET

SAMPLE DESCRIPTION

Calculation of Degradation
(% DOC Removal)

Date: 5-21-80
Analyst: W. 9. Schief
Hrs.:

FC-203 (day 7)

$D_{t_1} = \left[1 - \frac{37.4 - (-0.1)}{41.5 - 1.4}\right] \times 100 = 6.5\%$

$D_{t_2} = \left[1 - \frac{32.0 - (-0.1)}{41.6 - 1.4}\right] \times 100 = 20.1\%$

MEAN 13%

FC-203 (day 14)

$D_{t_1} = \left[1 - \frac{6.9 - 0.4}{40.1}\right] \times 100 = 83.8\%$

$D_{t_2} = \left[1 - \frac{6.0 - 0.4}{40.2}\right] \times 100 = 86.1\%$

85%

FC-203 (day 21)

$D_{t_1} = \left[1 - \frac{5.4 - 1.2}{40.1}\right] \times 100 = 89.5\%$

$D_{t_2} = \left[1 - \frac{2.4 - 1.2}{40.2}\right] \times 100 = 97.0\%$

93%

FC-203 (day 27)

$D_{t_1} = \left[1 - \frac{4.1 - 0.6}{40.1}\right] \times 100 = 91.3\%$

$D_{t_2} = \left[1 - \frac{4.2 - 0.6}{40.2}\right] \times 100 = 91.0$

91%

FC-203 (day 28)

$D_{t_1} = \left[1 - \frac{2.8 - 0}{40.1}\right] \times 100 = 93.0\%$

$D_{t_2} = \left[1 - \frac{2.8 - 0}{40.2}\right] \times 100 = 93.0\%$

93%

Reviewed by:

FORM 10620-8-PWO

File: Page No. 5541 S

ENVIRONMENTAL ENGR. LAB – WORK SHEET

SAMPLE DESCRIPTION

Calculations of Degradation (% DOC Removal)

Date: 5-22-80
Analyst: W.A. Schiff
Hrs.:

FC-3017

(day 7) $D_{t_1} = \left[1 - \frac{30.8 - (-0.1)}{41.1 - 1.4}\right] \times 100 = 22.2\%$ (39.7)

$D_{t_2} = \left[1 - \frac{19.2 - (-0.1)}{41.8 - 1.4}\right] \times 100 = 52.2\%$ (40.4)

MEAN 37%

FC-3017

(day 14) $D_{t_1} = \left[1 - \frac{2.4 - 0.4}{39.7}\right] \times 100 = 95.0\%$

$D_{t_2} = \left[1 - \frac{3.6 - 0.4}{40.4}\right] \times 100 = 92.1\%$

94%

FC-3017 (day 21) $D_{t_1} = \left[1 - \frac{3.0 - 1.2}{39.7}\right] \times 100 = 95.5\%$

$D_{t_2} = \left[1 - \frac{2.9 - 1.2}{40.4}\right] \times 100 = 95.8\%$

96%

FC-3017

(day 27) $D_{t_1} = \left[1 - \frac{2.5 - 0.6}{39.7}\right] \times 100 = 95.2\%$

$D_{t_2} = \left[1 - \frac{2.0 - 0.6}{40.4}\right] \times 100 = 96.5\%$

96%

FC-3017

(day 28) $D_{t_1} = \left[1 - \frac{1.5 - 0}{39.7}\right] \times 100 = 96.2\%$

$D_{t_2} = \left[1 - \frac{2.2 - 0}{40.4}\right] \times 100 = 94.6\%$

95%

Reviewed by:

FORM 10820-6-PWO

File: Page No. _____

ENVIRONMENTAL ENGR. LAB – WORK SHEET

SAMPLE DESCRIPTION Calculation of Degradation (% DOC Removal)

Date: 5-22-80
Analyst: W.A Schiff
Hrs.: _____

MEAN

FC-206 (day 7)

$D_{t_1} = \left[1 - \frac{31.0 - (-0.1)}{42.0 - 1.4}\right] \times 100 = 23.3\%$

$D_{t_2} = \left[1 - \frac{30.8 - (-0.1)}{41.8 - 1.4}\right] \times 100 = 23.5\%$

} 23%

FC-206 (day 14)

$D_{t_1} = \left[1 - \frac{5.7 - 0.4}{40.6}\right] \times 100 = 86.9\%$

$D_{t_2} = \left[1 - \frac{3.4 - 0.4}{40.4}\right] \times 100 = 92.6\%$

} 90%

FC-206 (day 21)

$D_{t_1} = \left[1 - \frac{3.6 - 1.2}{40.6}\right] \times 100 = 94.1\%$

$D_{t_2} = \left[1 - \frac{3.6 - 1.2}{40.4}\right] \times 100 = 94.1\%$

} 94%

FC-206 (day 27)

$D_{t_1} = \left[1 - \frac{3.4 - 0.6}{40.6}\right] \times 100 = 93.1\%$

$D_{t_2} = \left[1 - \frac{3.6 - 0.6}{40.4}\right] \times 100 = 92.6\%$

} 93%

FC-206 (day 28)

$D_{t_1} = \left[1 - \frac{3.5 - 0}{40.6}\right] \times 100 = 91.4\%$

$D_{t_2} = \left[1 - \frac{3.1 - 0}{40.4}\right] \times 100 = 92.3\%$

} 92%

Reviewed by: _____

FORM 10620-8-PWO

File: Page No. 5541 S

ENVIRONMENTAL ENGR. LAB – WORK SHEET

SAMPLE DESCRIPTION Calculation of Degradation (% DOC Removal)

Date: 5-23-80
Analyst: W. A Sieber
Hrs.:

MEAN

Hydroquinone (Day 7)

$D_{t_1} = \left[1 - \frac{2.0 - (-0.1)}{(17.3 - 1.4)}\right] \times 100 = 86.8\%$ (15.9)

$D_{t_2} = \left[1 - \frac{1.2 - (-0.1)}{17.0 - 1.4}\right] \times 100 = 91.7\%$ (15.6)

} 89%

Hydroquinone (Day 14)

$D_{t_1} = \left[1 - \frac{1.6 - 0.4}{15.9}\right] \times 100 = 92.5\%$

$D_{t_2} = \left[1 - \frac{1.8 - 0.4}{15.6}\right] \times 100 = 91.0\%$

} 92%

Hydroquinone (Day 21)

$D_{t_1} = \left[1 - \frac{2.7 - 1.2}{15.9}\right] \times 100 = 90.6$

$D_{t_2} \left[1 - \frac{0.8 - 1.2}{15.6}\right] \times 100 = (102.6\%)$ assuming 100% the mean = 95%

} 97%

Hydroquinone (Day 21)

$D_{t_1} = \left[1 - \frac{1.6 - 0.6}{15.9}\right] \times 100 = 93.7\%$

$D_{t_2} = \left[1 - \frac{1.4 - 0.6}{15.6}\right] \times 100 = 94.9\%$

} 94%

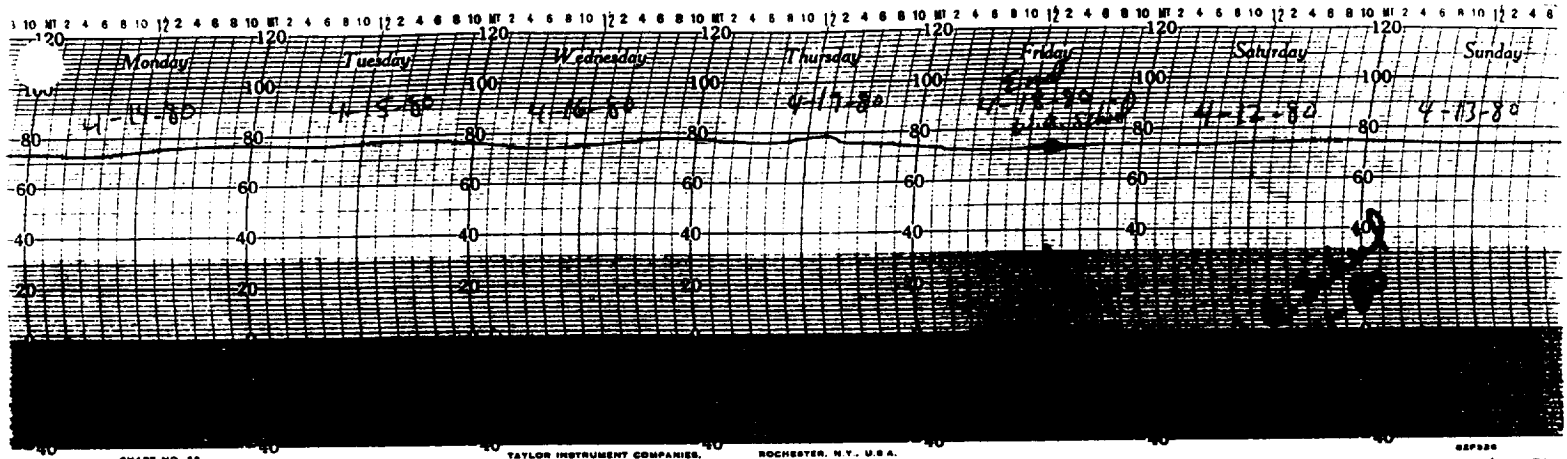
Hydroquinone (Day 28)

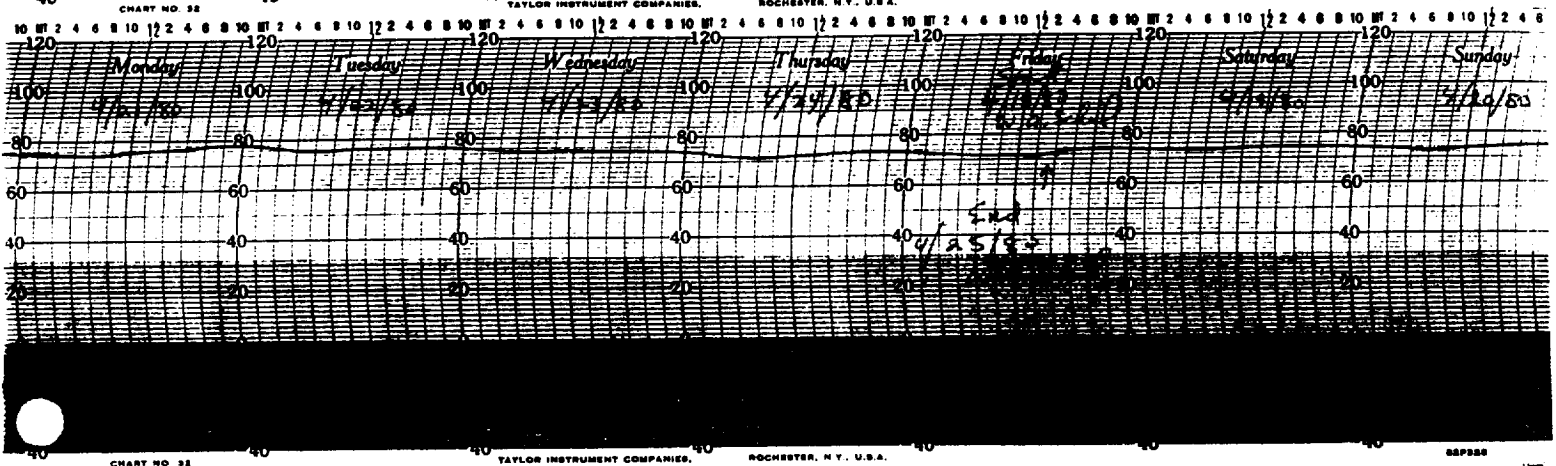
$D_{t_1} = \left[1 - \frac{1.4 - 0}{15.9}\right] \times 100 = 91.2\%$

$D_{t_2} = \left[1 - \frac{0.8 - 0}{15.6}\right] \times 100 = 94.9\%$

} 93%

Reviewed by:


Monday
Tuesday
Wednesday
Thursday
Friday
Saturday
Sunday
4-14-80
4-15-80
4-16-80
4-17-80
4-18-80
4-12-80
4-13-80
CHART NO. 32
TAYLOR INSTRUMENT COMPANIES, ROCHESTER, N.Y., U.S.A.


Monday
Tuesday
Wednesday
Thursday
Friday
Saturday
Sunday
4/21/80
4/22/80
4/23/80
4/24/80
4/25/80
4/19/80
4/20/80
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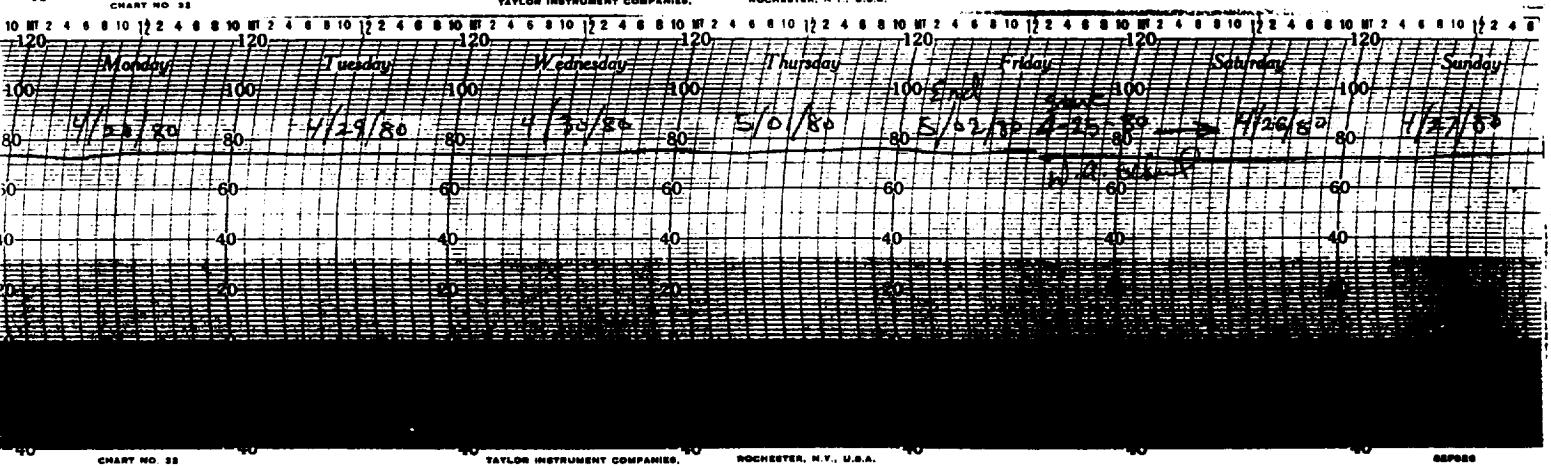
TAYLOR INSTRUMENT COMPANIES, ROCHESTER, N.Y., U.S.A.


Monday
Tuesday
Wednesday
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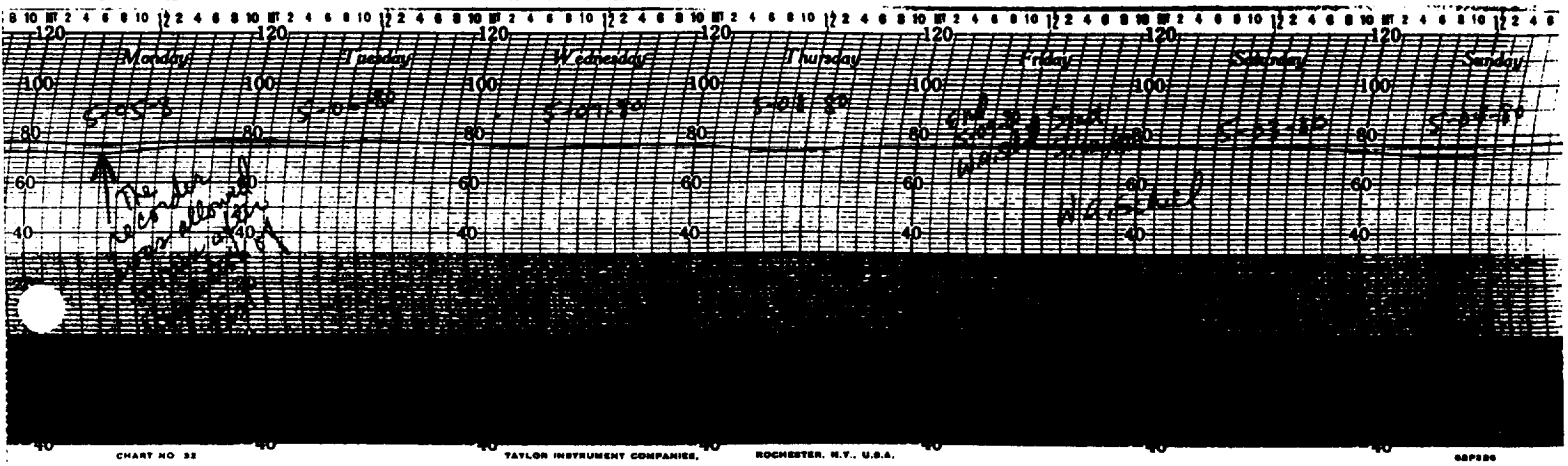
Saturday
Sunday
4/28/80
4/29/80
4/30/80
5/01/80
5/02/80
4/26/80
4/27/80
CHART NO. 32
TAYLOR INSTRUMENT COMPANIES, ROCHESTER, N.Y., U.S.A.


Monday
Tuesday
Wednesday
Thursday
Friday
Saturday
Sunday
5-05-80
5-06-80
5-07-80
5-08-80
5-04-80
5-03-80
CHART NO. 32
TAYLOR INSTRUMENT COMPANIES, ROCHESTER, N.Y., U.S.A.

FORM 10620-8-PWO

File: Page No. 5541 S

ENVIRONMENTAL ENGR. LAB – WORK SHEET

SAMPLE DESCRIPTION

Date: 5-22-80
Analyst: W. A. Schell
Hrs.:

Culture Medium Identification Code

sample number
x - (1) → duplicate dilutions
x - (2)
x - (3) — sterile control ($HgCl_2$ added)

Reviewed by:

Form 3337

LR NO. 5541-S

3M ENVIRONMENTAL LABORATORY DATA SHEET
MODIFIED OECD SCREENING TEST WITH DOC ANALYSIS
RESULTS OF DOC ANALYSES - EXPRESSED IN MG/L

FILE

SEC.

DATE

S

ANALYSIS FORM

	TEST DURATION	SAMPLE DESCRIPTION	BLANK C-1	BLANK C-2	BLANK C-3	ANTWERP FC-203, NFP 1-1	ANTWERP FC-203, NFP 1-2	ANTWERP FC-203, NFP 1-3	ANTWERP FC-3017, LOT-2303 2-1	ANTWERP FC-3017, LOT-2303 2-2	ANTWERP FC-3017, LOT-2303 2-3	ANTWERP FC-206 (FM-3834) LOT-2330 10/79 3-1	ANTWERP FC-206 (FM-3834) LOT-2330 10/79 3-2	ANTWERP FC-206 (FM-3834) LOT-2330 10/79 3-3	HYDROQUINONE (BIODEGRADABILITY STANDARD) 4-1	HYDROQUINONE (BIODEGRADABILITY STANDARD) 4-2	HYDROQUINONE (BIODEGRADABILITY STANDARD) 4-3
			A	B	C	E	F	G	I	J	K	M	N	O	Q	R	S
1	DAY - 0		1.1	1.6	1.2	42.4	44.0	40.3	41.4	42.3	40.3	42.3	42.3	40.8	17.1	17.8	17.7
2	(4-11-80)		1.9	1.2	1.5	40.6	39.8	45.1	40.8	41.2	41.6	41.7	41.3	42.0	16.3	16.2	15.0
3							41.1								18.4		10.8
4															16.2		
5															18.6		
6	DAY - 7		0.1	-0.1		37.0	31.8		31.2	18.4		31.0	30.9		2.3	2.6	
7	(4-18-80)		-0.4	0.1		39.1	32.1		30.3	19.9		31.0	30.7		1.7	0.6	
8						36.2										0.5	
9																	
10																	
11	DAY - 14		0.2	0.1		8.1	6.5		2.4	3.5		5.3	3.5		2.0	1.5	
12	(4-25-80)		0.5	0.6		6.6	5.5		2.5	3.8		6.1	3.3		1.2	2.4	
13						6.0										1.6	
14																	
15																	
16	DAY - 21		1.5	1.6	2.5	6.2	2.8	41.7	3.9	3.9		3.6	3.0		2.4	0.5	14.2
17	(5-02-80)		0.9	1.0	0.5	4.8	2.0		2.2	1.9		3.5	4.1		2.3	1.2	14.3
18					2.3	7.5									3.3		
19						4.2											
20						4.1											
21	DAY - 27		0.9	0.4		4.1	3.2		1.9	1.7		3.4	4.1		1.3	1.5	
22	(5-08-80)		0.1	0.8		4.1	5.0		3.2	2.2		3.4	3.2		1.9	1.4	
23							4.6		2.5								
24							4.2										
25																	
26	DAY - 28		0.1	-0.1	2.1	2.7	2.9	38.4	2.0	2.1	36.7	3.0	2.6	39.1	1.7	0.8	13.0
27	(5-09-80)		0.1	-0.1	1.7	2.8	2.6	40.8	1.0	2.4	36.9	4.5	3.6	41.6	1.0	0.8	14.2
28												3.0		38.1			13.7
29																	
30																	

THREE (3) REACTION VESSELS WERE FILLED WITH EACH TEST SOLUTION: EACH PORTION WAS INOCULATED.

REACTION VESSELS X-1 AND X-2 WERE KEPT AS DUPLICATES.

TO REACTION VESSEL X-3 WAS ADDED MERCURIC CHLORIDE FOR USE AS A STERILE CONTROL.