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Item ID Number	04090 ☐ **Not Scanned**
Author	
Corporate Author	
Report/Article Title	Foreign Correspondence on Requests for Tech Reports 1980 (USAF SAM/EK)
Journal/Book Title	
Year	0000
Month/Day	
Color	☐
Number of Images	28
Descripton Notes	Contents of folder originally labeled: "Foreign Correspondence on Requests for Tech Reports 1980 (USAF SAM/EK)."

- Comments / observations of 2 meetings -
- Reviewed the actions of Givaudan
  - Chemical Analysis Team
  - Medical team
  - Decontamination Team.
  Vaterlaus Briefed Authorities
- Offered to Decontaminate.
- Frustration
- Discussion other possible decont procedures - Biodegradation Rate
- Asked about metal.
- Discussed Volcanoes
- Significance to Givaudan
- TNO vs their lab. offered to share info

17 Nov 77

Givaudan Research Company

Dubendorf Switzerland

Conference Attendees: Young & Trembly

Dr Bruno Vaterlaus, Director of Research.

Dr. E. Homberger

Dr. H-K. Wipf.

Dr. Ralf Hütter, Prof. of Microbiology, Zurich University.

Dr. Viktor Krasnobajew

Dr. Norbert Neuner

A. Reviewed ICMSA Contamination Incident at Seveso, Italy 10 July 1976.
Dr Vaterlaus emphasized the preparations of Givaudan during the first after the incident

B. Reviewed Decontamination Program.

C. Inquired as to any additional Decont. Techniques

D. Discussed Our Data.

Microbiological Section

Josef Zeyer

Dr. Prof. Ralf Hütter

Martin Philippi

Dr. Viktor Krasnobajew

Decontamination

Biodegradation —

Groups investigation Biodegradation

1 {Prof. Negri - Dr. Ulc Pocchari}
2 Univ. of Milano
3 Huttinger, Amster.
4 Wattoc - Negri FEB-77

Preparation

1. Knowledge About Analytical Proc.
   Avoid Matsummura Spot
2. Extraction of labelled TCDD
3. Organisms Isolated.

*Send procedure.

EIDGENÖSSISCHE
TECHNISCHE HOCHSCHULE ZÜRICH

Mikrobiologisches Institut

CH-8006 Zürich, Universitätstrasse 2
Telephon 01-32 62 11

January 23, 1979

Dr. A.L. Young
Occupational and Environmental Health Laboratory
Brooks Air Force Base Texas
USA

Dear Dr. Young,

It is a long time, since we took contact with you. This is not due to any disinterest in the recent work on TCDD-biodegradation, but only to my personal laziness in writing.

As I hope you enjoyed your stay in Switzerland in November 1977 and the following trip to and partially through Italy. We certainly remember your visit here with pleasure. Will you again be in Europe in the near future?

As you know we have started a study on microbial metabolism of TCDD, in the hope to contribute to a solution of the Seveso-problem. The experimental work was started late in 1977 and will last until the end of 1979. The recent status is depicted in the added summary.

During our studies we encountered several problems, mostly analytical. I am sure, that some of the problems we are faced with are the same as you and your group have met. And if I take up my notes about our last discussions (nov. 17, 1977) I see that we also talked about this during your visit. May I take up some of these problems again?

a) The extraction of TCDD from soil can be performed with different solvent systems. You used methanol/chloroform. What were the reasons for this preference e.g. over methylen-chloride? Which ratio of methanol/chloroform was used, which concentrations, temperatures and extraction times?

b) Is it possible to get samples of the 15 2,4-D degrading strains, you mentioned during your last visit? What is known about the degradation pathway of these organisms? Do they only use the phenoxyacetic acid side chain as carbon source and leave the chlorinated ring or are they also capable of splitting the ring?

c) Can we get in contact with the researchers in the field of microbial laboratory biodegradation studies? If I remember correctly one name you mentioned was

Alsea, Oregon

Testimony is to address the environmental fate of TCDD. I have attempted to examine four major questions germane to the environmental fate of TCDD:

1. Does TCDD persist in the environment when sprayed as a contaminant of 2,4,5-T herbicide?

2. Does TCDD move in the abiotic (non-living) portion of the environment?

3. Does TCDD bioaccumulate or biomagnify within biological organisms in the field environment or their food (crops/animals/plants)?

4. Does TCDD have an adverse effect upon the organisms in which it is found in the environment?

Do the levels of TCDD found in the environment have adverse effect upon the organisms within that environment

AF Legal Counsel

Mr Bill Morrison

Rm 4C-927

PENTAGON

Washington D.C.

225-4691

Absence of TCDD Toxicity to a Rodent Population Following Massive Field Application of 2,4,5-T Herbicide*

Charles E. Thalken, DVM, MS; William E. Ward, PhD

Alvin L. Young, PhD

USAF Academy, Colorado

Field investigations were conducted on populations of beach mice, Peromyscus polionotus, and hispid cotton rats, Sigmodon hispidus from a unique 1 square mile military test site (Test Area C-52A, Eglin AFB, Florida) that was sprayed with 160,948 pounds of active ingredient 2,4,5-trichlorophenoxyacetic acid herbicide (2,4,5-T). Significant levels (10-710 parts per trillion - ppt) of the contaminant 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) were found within the top 6 inches of test site soils although 10 years had elapsed since the last aerial application of 2,4,5-T. Liver tissue from rodents inhabiting the test site contained 210-1,300 ppt TCDD. However, no gross or histological evidence of teratogenesis or toxicity was found in 122 adults and 87 fetuses. An analysis of variance of liver and spleen weights for the beach mouse indicated significant differences between control and TCDD-exposed animals. Analysis of plant seeds revealed no detectable levels of TCDD (minimum detection limit of 1 ppt TCDD). TCDD accumulation in liver tissue was thought to be associated with pelt contamination from burrowing and subsequent ingestion of soil particles via grooming.

* Presentation to the American Veterinary Medical Association, 112th Annual Meeting, Anaheim, California, 16 July 1975, Abstract Number 81.

Dr. Arnold, another Dr. Cairney. Is this correct?

d) In the USAF-report the dissappearance of TCDD from the experimental fields is reported. The extensive studies on the Eglin Air Force Base (Florida) are especially interesting, as they give the well known "half life" of 300 days, the same as in Utah. Is it possible to get soil samples from Eglin Air Force Base, especially from the location with originally heavy contamination (10-40 ppm TCDD). To whom will we have to address such a request?

We realize that answering some of the questions may be time consuming and difficult, especially as you are working in different fields now. But we would be very grateful, if you could at least give us the address of the research microbiologists at Colorado Springs and an address for point d). In advance we thank you for all your help.

Yours sincerely,

Ralf Hütter

R. Hütter

BEILAGE 2
Zu Bericht über Besprechung in Seveso vom 10.1.1979

EIDGENÖSSISCHE
TECHNISCHE HOCHSCHULE ZÜRICH

Mikrobiologisches Institut

CH-8006 Zürich, December 22, 1978
Universitätstrasse 2
Telephon 01-32 62 11

"Biodegradation of TCDD"? A Swiss Study of the Problem.

(M. Philippi and R. Hütter, Mikrobiologisches Institut, ETH-Zentrum, 8092 Zürich)

In Spring 1977 we were asked by the Swiss Government (Department of External Affairs) and by Givaudan Co. to study the possibility of microbial degradation of TCDD. After some time, used to inform ourselves on the situation in Seveso and on earlier or on-going work in other laboratories, a proposition for an experimental programme was submitted to the Swiss Government. The proposal was accepted late in 1977.

The programme is financed by the Swiss Federal Government, and one of us (R.H.) is commissioned to direct it. The experimental work (performed exclusively by M.P.) is done in a special laboratory in the research buildings of Givaudan Co. in Dübendorf, a suburb of Zürich. This arrangement had to be made for reasons of safety (TCDD work should not be done in student laboratories) and for analytical reasons (all facilities and experienced personel are available in the same building).

More than 100 different microorganisms, single cultures and mixed populations, are examined under various culture conditions for their capacity to attack TCDD. ^{14}C-TCDD, obtained from Kor Isotopes, is used for the studies.

In addition to work in our own laboratory we are cooperating with Prof. Salkinoja, University of Helsinki, in a study of TCDD-degradation by "Biox"-filters, patented by Enso-Gutzeit Co., Finnland.

The incubations are made in closed systems, thus allowing a complete analysis of the fate of TCDD. The actual analysis permits the detection of labelled CO_2 by trapping it from the outflowing air in KOH. Hydroxylated metabolites should be detected in thin layer chromatograms; they should occur primarily in the water phase after extraction of the incubated media with organic solvents. We analyse for partially or completely dechlorinated metabolites in a GC-system.

Until present no development of $^{14}CO_2$ could be detected in any culture. All potential polar metabolites, observed in the water phase after extraction with organic solvents and as radioactive spots remaining at the start in thin layer chromatograms, turned out to be artefacts. They were due to incomplete extraction of TCDD, which seems to be especially strongly bound to cell particles in some fungal cultures.

Also the analysis of the samples in the GC-system did, until present, not lead to any detectable TCDD-metabolite. A new peak, detected in 2 cultures after several months of incubation, did cochromatograph with Monchlor-DD, but turned out later not to be radioactively labelled. As this compound can therefore not be a metabolite of the ^{14}C-TCDD added to the culture, its identity was not investigated further.

Recently we have started with an analysis of our samples in a Packard Radio-GC-System. With this system it is possible to analyze a sample simultaneously in a GC-detector and a radioactivity proportional counter. In preliminary experiments 2 samples were found which contain a new peak of radioactively labelled material not present in the standard.

Future work is planned in the following directions:

- Further analysis of the two samples showing a new peak in the Radio-GC-System.
- Analysis of the remaining samples in the Radio-GC-System.
- Complete analysis of all samples incubated for at least 1 year (to determine the balance of the added radioactivity).
- Start new set of microbial cultures under modified conditions.

R Hutter

EIDGENÖSSISCHE
TECHNISCHE HOCHSCHULE ZÜRICH
Mikrobiologisches Institut
Tel. 01. 92 62 11

CH-8006 Zürich,
Universitätstrasse 2

Dear Doctor Young

as we hope you received
our last letter (january 23).
Could you please communicate
to us the address, to whom
we will have to address our
request ?
Looking forward to your
answer,

Yours sincerely

R. Hütter

January 23, 1979

copy agn 8, 1979

Mikrobiologisches Institut
Eidg. Techn. Hochschule
ETH-Zentrum
8092 Zürich

Dr. A.L. Young
Occupational and Environmental Health Laboratory
Brooks Air Force Base Texas
USA

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Yours sincerely,

R. Hütter

9 April 1979

Dr. Ralf Hutter
Mikrobiologisches Institut
Eidgenossische Technische Hochschule
CH - 8006, Zurich Switzerland

Dear Dr Hutter

Thank you for your letters of 23 Jan and 8 Apr 79. I apologize for my delay in replying to your letters. Hocutt's Significant events related to the phenoxy herbicides and TCDD occur almost weekly and my time is spent on field trips or engaged in extensive report preparations.

Your hospitality and interest in my research were most appreciated during my visit to Zurich. I hope to visit Europe next year. I am most interested in the results of your work. I have continued to read reports on Seveso and the data appear not to be encouraging. First to answer your questions posed in your letters:

a. Extraction systems for TCDD: We experimented with a number of extraction systems for TCDD. The methanol/chloroform system was one that we

d. Eglin Air Force Base, Florida Studies: Table 2 contains recent data on the persistence of TCDD in soils of the Eglin Test Area. As you can see, in this area, where the TCDD is no longer associated with the chlorophenoxy herbicides, the half-life appears to have increased to approximately three years (assuming that sampling and analytical variations permit valid means). Soils of the test area (now containing approximately 320 ng TCDD/kg soil) are available + I have asked my colleague at Eglin to collect and forward a soil sample to you immediately. I will not have analyzed the specific sample, but would be interested in your data on its TCDD content.

We are now proposing some new decontamination studies for the areas where Herbicide Orange was stored prior to its incineration in the fall of 1977. Chemical and radiological techniques have been proposed and some initial laboratory data obtained. I'm no longer optimistic about the use of microbial strains to enhance TCDD degradation, although we certainly haven't ruled them out.

Extraction times were 30 minutes at 60 °C. * temperature

microbial media

used in our earlier studies. Others we have tried included a 5 percent mixture of dichlorobenzene in chloroform and a mixture 10 percent toluene in chloroform. We obviously selected these solvent systems because of the solubility of TCDD. However, if I correctly understand your question, you are concerned with TCDD from soil rather than culture media. Our analytical group here found that - - - -

b. Microbial Strains: I still have access to the fifteen strains that I mentioned to you during our visit. However we have found that individual strains to be ineffective in degrading TCDD. They were especially Aspergillus Ochraceus, capable of rapid degrading 2,4-D and 2,4,5-T. Dr William J. Cairney has maintained these cultures and will provide them to you upon request. However I believe them to be of little value to you.

both chain and ring degradation

C. Degradation Studies: Dr Cameron (Department of Chemistry and Biological Sciences, United States Air Force Academy, Colorado U.S.A. 80840) and I have continued our studies on the Herbicide Biodegradation plots and on areas at Gulfport, Mississippi where spills of 2,4-D and 2,4,5-T occurred. The collection of data has been slow and tedious. I have attached a recent ~~one~~ report from Dr Cameron on the microbial studies. Two dominant organisms (a yeast, Geotrichum sp. and a fungus, Pseudomonas fluorescens) have been isolated from the study areas at Gulfport, Mississippi, that appear to have a close association with high levels of herbicides & TCDD ~~degradation~~. Table 1 presents our most recent data (reported 7 MAY 79) on levels of herbicides and TCDD on these spill areas. I will ask Dr Cameron to furnish you with ~~either "fresh"~~ cultures of the two organisms in question.

we have two significant reports in preparation. I do not believe however that they will be released prior to August. I will forward them to you upon their release.

I am sending under separate cover a copy of my recent technical report on Herbicide Orange (Technical Report OEHL-TR-078.92). Again please accept my apology for delay in correspondence and express my sincere regards to , Dr Hans Wiff and Dr Vatenburg.

Trip Report

Attendance

- Jeremy J. Stone — F. A. S. (22 in Room)
- Packs
- Stellman, MS E.
- Tung
- Poole } EPA
- Preston }
- Marlowe (Penthouse)
- Steven Greg > Vietnam Vet Advisor
- Nicolas Wade - (Science)
- Steven Woodward - FDA

Confusion over DOSE vs Exposure

Briefed Sen D.

Maj Chet Paul
LTCol Paul Fallon
Maj Phil Brown

AUTHOR'S COPY

Maj Young/EC/3667/11/30 May 79/5-30-1

31 MAY 1979

EC

Literature Request

Mr Elbert L. Dage
Office of Toxic Substances (TS-792)
Assessment Division
Environmental Protection Agency
Washington, D.C. 20460

1. Per your request for literature, the following documents on Air Force studies of the environmental fate of 2,4-D, 2,4,5-T and TCDD are attached.

2. The first three Technical Reports are pertinent studies on the fate of 2,4,5-T and its trace contaminant TCDD in an ecosystem treated with massive quantities of phenoxy herbicides. Technical Reports AFATL-TR-74-12 (Atch 1), AFATL-TR-75-49 (Atch 2) and AFATL-TR-75-142 (Atch 3) detail ecological studies conducted on a unique 3.0 km^2 military test area (Test Area C-52A, Eglin AFB FL) that received approximately 73,000 kg 2,4,5-T and 77,000 kg 2,4-D during the period 1962-70. Significant results included:

a. At the termination of spray equipment testing programs in 1970, significant levels (ppm) of 2,4,5-T soil residues were found throughout the test area. However, no residues of 2,4,5-T were detected (detection limit of 10 ppb) in any soil samples collected during 1971-1972.

b. During the years from 1974 through 1978, 54 soil samples were collected to a depth of 15 cm from throughout the test area and analyzed for TCDD. TCDD levels ranged from <10 to 1,500 parts per trillion (ppt). The median concentration was 30 ppt while the mean was 165 ppt.

c. An ecological survey extending over a five-year period (1973-1978) documented the presence of more than a 123 different plant species, 77 bird species, 71 insect families, 20 species of fish, 18 species of reptiles, 18 species of mammals, 12 species of amphibians and 2 species of molluscs. At least 170 biological samples were analyzed for TCDD, including 30 species of animals. No TCDD was found in any of the plant species examined. However, TCDD was found in nine species of animals including two rodent species: beachmouse (300-1,500 ppt, liver) and hispid cotton rat (<10-210 ppt, liver); three species of birds: meadowlark (100-1,020 ppt, liver), mourning dove (50 ppt, liver), and Savannah sparrows (69 ppt, liver); three species of fish: spotted sunfish (85 ppt, liver) mosquitofish (12 ppt, whole body) and sailfin shiner (12 ppt, whole body), and one reptile, the six-lined racerunner (360-430 ppt, muscle). A composite sample of insects (whole bodies) contained 40 ppt TCDD.

d. Gross pathology was done on all species collected for TCDD residue analyses. Histopathological examinations were performed on over 300 beachmice or hispid cotton rats from the test area and a control field site. Examinations were performed on the heart, lungs, trachea, salivary glands, thymus, liver, kidneys, stomach, pancreas, adrenals, large and small intestine, spleen, genital organs, bone, bone marrow, skin and brain. Initially, the tissues were examined on a random basis without the knowledge of whether the animal was from a control or test area. All microscopic changes were recorded including those interpreted as minor or insignificant. The tissues were then reexamined on a control and test basis, which demonstrated that the test and control mice could not be distinguished histopathologically. Similar histopathological studies were conducted on the fish and racerunners with no significant abnormalities being found.

3. Technical Report USAFA-TR-76-18 (Atch 4) is a summary of Air Force ecological research on TCDD. It also presents analytical data on the soil degradation of TCDD when in the presence of 2,4,5-T and 2,4-D. Significant results included:

a. The half-life of TCDD in soils containing 2,4-D and 2,4,5-T appeared to be between 225 and 275 days.

b. Studies of bacteria, actinomycetes and fungi from soil plots treated with massive quantities of 2,4-D and 2,4,5-T (5,000 - 40,000 ppm) confirm that these microorganisms proliferate to such an extent that they were probably using the herbicides and TCDD as metabolic carbon sources and, as such, were contributing to their degradation.

c. Movement of TCDD in the abiotic portions of the environment occurred by wind or water erosion of soil particles, but leaching by water alone did not occur.

4. Technical Report USAF OEHL-TR-78-92 (Atch 5) contains, in addition to the above environmental data on 2,4,5-T and TCDD (Chapter III), other significant data including:

a. Data on the analysis of 492 samples of Herbicide Orange (a 50:50 mix of the n-butyl esters of 2,4-D and 2,4,5-T) for TCDD (See Chapter I). The levels of TCDD ranged from <0.02 to 15 ppm in herbicide produced during the 1965-1968 time period. The weighted mean concentrations of TCDD in Herbicide Orange was 1.98 ppm. The samples were taken from surplus Herbicide Orange.

b. Industrial hygiene and ambient air sampling data from all land-based dedrumming/transfer operations of Project PACER HO, the 1977 USAF project to dispose of 2.22 million gallons of Herbicide Orange (see Chapter II). Results of these sampling programs revealed that under the worst case noted, the levels of 2,4,5-T (and 2,4-D) vapors were well below the time-weighted Threshold Limit Value (TLV) for each of these materials. The detected levels were at least two and, in most cases, three orders of magnitude below the TLVs. TCDD was not detected in any air samples. Approximately 200 personnel carried out the dedrumming/transfer

operations. Comparisons of available pre- and post-operational medical examinations of military personnel involved have revealed no apparent physical effects as a result of these activities.

c. An assessment of the world's scientific literature on the toxicity of 2,4,5-T, 2,4-D and TCDD in selected laboratory and domestic animal species (see Chapter IV). Each chemical was critically reviewed for:

(1) Acute and short-term toxicity potentials.

(2) Subacute and chronic toxicity potentials.

(3) Absorption, distribution, and excretion potentials.

(4) Embryotoxic, fetotoxic, and teratogenic potentials.

(5) Carcinogenic and tumorigenic potentials.

(6) Mutagenic and cytogenic potentials.

d. A review of available scientific data on numerous incidents involving suspected 2,4,5-T/TCDD poisoning of humans or livestock (see Chapter V). Extensive efforts in translating over 30 major foreign documents permitted for the first time detailed accounts of 23 industrial episodes that involved exposure of over 1,100 people to TCDD. An assessment of the medical data from these industrial episodes and other episodes was made in Chapter VI. Some of the significant medical conclusions were:

(1) Adverse effects of 2,4,5-T herbicide should manifest themselves shortly after exposure. Symptoms arising for the first time months to years after the last exposure are probably due to an etiology other than the herbicide.

(2) The hallmark of TCDD exposure to chlorance and its absence makes it unlikely that systemic disorders present are related to TCDD. Asthenic and vegetative symptoms are often present in overexposure but are difficult to interpret. They would normally be expected to clear with time.

(3) There is no conclusive evidence at this time that either 2,4,5-T herbicide or TCDD is mutagenic, teratogenic, or carcinogenic in man.

5. If I can provide additional copies of these documents or further elaborate on the data, please contact me.

ORIGINAL SIGNED BY

ALVIN L. YOUNG, Major, USAF, Ph.D.
Environmental Sciences Consultant
USAF Herbicide Specialist

5 Atchs
1. AFATL-TR-74-12
2. AFATL-TR-75-49
3. AFATL-TR-75-142
4. USAFA-TR-76-18
5. USAF OEHL-TR-78-92

EIDGENÖSSISCHE
TECHNISCHE HOCHSCHULE ZÜRICH

Mikrobiologisches Institut

CH-8006 Zürich,
Universitätstrasse 2
Telephon 01-32 62 11

March 10, 1980

Major A.L. Young, USAF, Ph.D.
Department of the Air Force
School of Air Space Medicine SAM/EK
Brooks Air Force Base, Texas 78235
U.S.A.

Dear Major Young,

Quite some time has passed, since we have been in contact in connection with your Agent Orange and TCDD studies. As you may remember, we initiated a laboratory programme to study the possibility of microbial metabolism of TCDD. Our present data are purely negative. We find no degradation or metabolism of TCDD by any one of our microorganisms or in any one of the soil samples studied up to now (we can not exclude a 1-2 % metabolism within 1 year). However many different test artefacts occur, e.g. overall diminuition of extractability, selective extraction of contaminants present in the ^{14}C-TCDD standard or seemingly polar metabolites.

As Dr. D. Watkins, EPA, Cincinatti, told me you are still interested and engaged in TCDD (or Agent Orange) work. We would be especially interested, whether your studies on the disappearance of TCDD from the soil of different experimental fields were continued and whether the "half-life" of about 1 year is still valid. From our laboratory data and from field experiments at Seveso, Italy, a longer half-life is suggested. Are any studies continued?

In advance we thank you for any information or help which you can give us.

Yours sincerely,

R. Hütter

8 April 1980

Dr. Ralf Hütter
Eidgenössische
Technische Hochschule Zürich
Mikrobiologisches Institut
Universitätstrasse 2
8006 ZURICH SWITZERLAND

Dear Dr. Hütter

Thank you for your letter of 10 March 1980. I apologize for my delays in corresponding with you. Significant events related to the phenoxy herbicides and TCDD continue to dominate my time.

Your hospitality and interest in my research were most appreciated during my visit to Zurich. Following my return from Europe, I transferred to the United States Air Force Occupational and Environmental Health Laboratory and the School of Aerospace Medicine, Brooks AFB, San Antonio, Texas. In this post, I have continued my investigations into the fate of TCDD. As you are aware, investigations of TCDD are extremely difficult and time consuming. We have not had the opportunity to extend our investigations beyond our field studies; i.e., we have not undertaken extensive microbial degradation studies in the laboratory. Hence, our observations must be viewed with caution as to cause and effect.

One of our major problems (and apparently one of yours) has been finding an efficient extraction technique to remove TCDD from soil. I believe our earlier data reflected this problem. With time, it may well be that TCDD becomes more tightly bound into the soil matrix and less extractable. Thus, our earlier data may have reflected binding and not degradation. We have experimented with a number of extraction techniques including exhaustive hexane extraction, methanol/chloroform, five percent dichlorobenzene in chloroform and a mixture of ten percent toluene in chloroform. Perhaps our most consistent method and the one used in obtaining the data in Technical Report OEHL-TR-79-169 is as follows:

> Ten grams of soil were weighed in a 6-dram vial. Six ml of caustic methanol (10% KOH, w/v) were added and shaken at 56°C overnight to hydrolyze the herbicide components to water-soluble products. Six ml of water were added and the vial shaken vigorously to break up any soil which had caked in the strongly basic solution. The sample was then extracted by the

addition of three ml of hexane and one and one-half ml of the supernatant liquid was pipetted into a two ml crimp-top vial. The hexane extract was sealed in this vial for direct GC/MS analysis. Samples with less than 50 ppb TCDD were further concentrated by careful air evaporation of the hexane to leave residues which were then redissolved in the desired amount of solvent. This latter step allowed for up to two orders of magnitude concentration of the sample over the levels present in the original one and one-half ml extract, and permitted the detection of 0.1 ppb in the soil sample with a signal to noise ratio greater than 3:1.

I have attached copies of two recent reports. The first (Atch 1) is my presentation to the American Chemical Society, 14 Sep 79. Of particular interest in Vugraph 6 (pg 14) on the disappearance of TCDD from soils collected in 1974 and 1978 from Eglin AFB, Florida. These data suggest that the half-life of TCDD may be in excess of three years. However, the data show considerable variation and I hesitate to conclude very much from them. The important point to note is that this area (Grid I) received the TCDD in the 1962-64 time period and as of 1971-72, no detectable levels of 2,4-D or 2,4,5-T were found in these soils. The second report (Atch 2) is on our studies at Gulfport, Mississippi. From this location we have approximately two years of what I believe are good data. The levels of herbicides, phenols and TCDD are high. We have noted a decrease in TCDD in these soils but our data over time do not yet permit an accurate determination of half-life (certainly it will exceed two, perhaps three years). Tables 3,4 and 5 (pg 18, 19 and 22) of this second report show some important data. As noted we are seeing some fairly high levels of microorganisms in the plots with high levels of herbicide and TCDD. The collection of these data has been slow and tedious. Dr. Cairney (Major William J. Cairney, Ph.D., Department of Chemistry and Biological Sciences, USAF Academy, Colorado 80840, USA) has continued to isolate and identify microbial organisms from the TCDD-contaminated soils. The value of these organisms in degrading TCDD in soil substrates has not been evaluated. I recommend you contact Dr. Cairney for isolate information. I've attached (Atch 3) one of Dr. Cairney's in-house reports on microbial analyses of soils from our Herbicide Orange Biodegradation Plots.

We have proposed some decontamination studies for the sites at Gulfport, Mississippi and Johnston Island, Pacific Ocean where Herbicide Orange was stored prior to its incineration in the summer of 1977. Chemical and radiological techniques have been proposed and we have already obtained some initial laboratory data. I'm no longer optimistic about the use of microbial strains to enhance TCDD degradation, although we certainly haven't ruled them out. We have a report in preparation on our Johnston Island studies. I will forward it to you when it is available.

Again, please accept my apologies for the long delay in correspondence. Please express my sincere regards to Mr. Josef Zeyer and Drs Wipf, Neuner and Vaterlaus.

Sincerely

ALVIN L. YOUNG, Major, USAF, Ph.D.
Consultant, Environmental Sciences

3 Atch
1. American Chemical Society Presentation
2. OEHL-TR-79-169
3. Microfloral Analyses of Soil from Phenoxy Herbicide Biodegradation Plots at Eglin AFB, Florida, and the AFLC Test Range, Utah

Cy to: Dr David Watkins
US EPA
26 West St. Clair
Cincinnati OH 45268
w/atch

EC

Literature Request

1. Per your request for literature, the following documents on Air Force studies of the environmental fate of 2,4-D, 2,4,5-T and TCDD are attached.

2. The first three Technical Reports are pertinent studies on the fate of 2,4,5-T and its trace contaminant TCDD in an ecosystem treated with massive quantities of phenoxy herbicides. Technical Reports AFATL-TR-74-12 (Atch 1), AFATL-TR-75-49 (Atch 2) and AFATL-TR-75-142 (Atch 3) detail ecological studies conducted on a unique 3.0 km^2 military test area (Test Area C-52A, Eglin AFB FL) that received approximately 73,000 kg 2,4,5-T and 77,000 kg 2,4-D during the period 1962-70. Significant results included:

a. At the termination of spray equipment testing programs in 1970, significant levels (ppm) of 2,4,5-T soil residues were found throughout the test area. However, no residues of 2,4,5-T were detected (detection limit of 10 ppb) in any soil samples collected during 1971-1972.

b. During the years from 1974 through 1978, 54 ~~Fifty-four (54)~~ soil samples were collected to a depth of 15 cm from throughout the test area and analyzed for TCDD. TCDD levels ranged from <10 to 1,500 parts per trillion (ppt). The median concentration was 30 ppt while the mean was 165 ppt.

c. An ecological survey extending over a five-year period (1973-1978) documented the presence of more than a 123 different plant species, 77 bird species, 71 insect families, 20 species of fish, 18 species of reptiles, 18 species of mammals, 12 species of amphibians and 2 species of molluscs. At least 170 biological samples were analyzed for TCDD, including 30 species of animals. No TCDD was found in any of the plant species examined. However, TCDD was found in nine species of animals including two rodent species: beachmouse (300-1,500 ppt, liver) and hispid cotton rat (<10-210 ppt, liver); three species of birds: meadowlark (100-1,020 ppt, liver), mourning dove (50 ppt, liver), and Savannah sparrows (69 ppt, liver); three species of fish: spotted sunfish (85 ppt, liver) mosquitofish (12 ppt, whole body) and sailfin shiner (12 ppt, whole body), and one reptile, the six-lined racerunner (360-430 ppt, muscle). A composite sample of insects (whole bodies) contained 40 ppt TCDD.

d. Gross pathology was done on all species collected for TCDD residue analyses. Histopathological examinations were performed on over 300 beachmice or hispid cotton rats from the test area and a control field site. Examinations were performed on the heart, lungs, trachea, salivary glands, thymus, liver, kidneys, stomach, pancreas, adrenals, large and small intestine, spleen, genital organs, bone, bone marrow, skin and brain. Initially, the tissues were examined on a random basis without the knowledge of whether the animal was from a control or test area. All microscopic changes were recorded including those interpreted as minor or insignificant. The tissues were then reexamined on a control and test basis, which demonstrated that the test and control mice could not be distinguished histopathologically. Similar histopathological studies were conducted on the fish and racerunners with no significant abnormalities being found.

3. Technical Report USAFA-TR-76-18 (Atch 4) is a summary of Air Force ecological research on TCDD. It also presents analytical data on the soil degradation of TCDD when in the presence of 2,4,5-T and 2,4-D. Significant results included:

a. The half-life of TCDD in soils containing 2,4-D and 2,4,5-T appeared to be between 225 and 275 days.

b. Studies of bacteria, actinomycetes and fungi from soil plots treated with massive quantities of 2,4-D and 2,4,5-T (5,000 - 40,000 ppm) confirm that these microorganisms proliferate to such an extent that they were probably using the herbicides and TCDD as metabolic carbon sources and, as such, were contributing to their degradation.

c. Movement of TCDD in the abiotic portions of the environment occurred by wind or water erosion of soil particles, but leaching by water alone did not occur.

4. Technical Report USAF OEHL-TR-78-92 (Atch 5) contains, in addition to the above environmental data on 2,4,5-T and TCDD (Chapter III), other significant data including:

a. Data on the analysis of 492 samples of Herbicide Orange (a 50:50 mix of the n-butyl esters of 2,4-D and 2,4,5-T) for TCDD (See Chapter I). The levels of TCDD ranged from <0.02 to 15 ppm in herbicide produced during the 1965-1968 time period. The weighted mean concentrations of TCDD in Herbicide Orange was 1.98 ppm. The samples were taken from surplus Herbicide Orange.

b. Industrial hygiene and ambient air sampling data from all landbased dedrumming/transfer operations of Project PACER HO, the 1977 USAF project to dispose of 2.22 million gallons of Herbicide Orange (see Chapter II). Results of these sampling programs revealed that under the worst case noted, the levels of 2,4,5-T (and 2,4-D) vapors were well below the time-weighted Threshold Limit Value (TLV) for each of these materials. The detected levels were at least two and, in most cases, three orders of magnitude below the TLVs. TCDD was not detected in any air samples. Approximately 200 personnel carried out the dedrumming/transfer

operations. Comparisons of available pre- and post-operational medical examinations of military personnel involved have revealed no apparent physical effects as a result of these activities.

c. An assessment of the world's scientific literature on the toxicity of 2,4,5-T, 2,4-D and TCDD in selected laboratory and domestic animal species (see Chapter IV). Each chemical was critically reviewed for:

1. Acute and short-term toxicity potentials.

2. Subacute and chronic toxicity potentials.

3. Absorption, distribution, and excretion potentials.

4. Embryotoxic, fetotoxic, and teratogenic potentials.

5. Carcinogenic and tumorigenic potentials.

6. Mutagenic and cytogenic potentials.

d. A review of available scientific data on numerous incidents involving suspected 2,4,5-T/TCDD poisoning of humans or livestock (see Chapter V). Extensive efforts in translating over 30 major foreign documents permitted for the first time detailed accounts of 23 industrial episodes that involved exposure of over 1,100 people to TCDD. An assessment of the medical data from these industrial episodes and other episodes was made in Chapter VI. Some of the significant medical conclusions were:

1. Adverse effects of 2,4,5-T herbicide should manifest themselves shortly after exposure. Symptoms arising for the first time months to years after the last exposure are probably due to an etiology other than the herbicide.

2. The hallmark of TCDD exposure is chloracne and its absence makes it unlikely that systemic disorders present are related to TCDD. Asthenic and vegetative symptoms are often present in overexposure but are difficult to interpret. They would normally be expected to clear with time.

3. There is no conclusive evidence at this time that either 2,4,5-T herbicide or TCDD is mutagenic, teratogenic, or carcinogenic in man.

5. If I can provide additional copies of these documents or further elaborate on the data, please contact me.

MAJOR

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Environmental Sciences Consultant
USAF Herbicide Specialist

5 Atchs
1. AFATL-TR-74-12
2. AFATL-TR-75-49
3. AFATL-TR-75-142
4. USAFA-TR-76-18
5. USAF OEHL-TR-78-92