METABOLISM AND PHARMACOKINETICS OF HALON 1211 AND ITS POTENTIAL REPLACEMENTS HCFC-123 AND PERFLUOROHEXANE

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The animals used in this study were handled in accordance with the principles stated in the Guide for the Care and Use of Laboratory Animals, prepared by the Committee on Care and Uses of Laboratory **Animals** of the Institute of Laboratory Animal Resources, National Research Council, DHHS, National Institute of Health Publication Number 86-23, 1985, and the Animal Welfare Act **of** 1966, as amended.

INTRODUCI'ION

Halon 1211 currently is being used by the US Air Force as a flight line fire extinguishant. Due to environmental concerns of ozone depletion, Halon 1211 must be phased out by the year 2000. Before a replacement candidate can be chosen, fire suppression ability, environmental concerns, and the toxicity of prospective candidates need to be considered.

This study has investigated the metabolism and pharmacokinetics of HCFC-123, perfluorohexane, and Halon 1211 in Fischer 344 and Sprague Dawley rats. Structures of the compounds studied are shown in Figure 1. The objective of the metabolism study was to find metabolites of toxicological significance. For the metabolism studies animals were exposed via nose only inhalation to a 1% atmosphere of the test material for **2** hours. Tissues were analyzed for volatile and nonvolatile metabolites. Urine was analyzed for fluoride, bromide, and nonvolatile metabolites. **For** the pharmacokinetic studies animals were either anesthetized, tracheotomized and ventilated **from** mylar bags containing varying concentrations of the test material or exposed in inhalation chambers. Expired breath, sampled from the tracheotomized animals, was analyzed for parent compound and volatile metabolites. Blood was analyzed for parent compound and metabolites and urine was analyzed for metabolites. Data from pharmacokinetic studies, and gas uptake studies were used to construct a physiological based pharmacokinetic (PBPK) model describing the metabolism of HCFC-123.

METHODS

ANIMALS

Metabolism

Male Fischer 344 and Sprague Dawley rats were exposed to a 1% atmosphere of test material for 2 h via nose only inhalation. The Fisher 344 rats were 8 weeks old weighing 150-200 g and the Sprague Dawley rats were 6 weeks old weighing 175-225 g. Groups of 8 animals and 2 controls were used for each test material. Control animals were exposed to air only using the same apparatus. Immediately after exposure 4 test animals and 1 control animal were sacrificed. Samples of liver, kidney, heart, lung, muscle, skin, fat, testes, and blood were quick-frozen in liquid nitrogen. The remaining 4 test animals and 1 control animal were put into metabolism cages for 24 h urine and feces collection. After 24 h the 4 test animals and 1 control animal were sacrificed. Tissue samples were obtained for analysis. A flow diagram of the inhalation exposure experiments is shown in Figure 2.

Pharmacokinetics

Male Fischer 344 rats were exposed to 1.0%, 0.1%, or 0.01% of HCFC-123 for a period of 4 h. Animals were either anesthetized, tracheotomized, and ventilated artificially from mylar bags containing the exposure atmosphere or were awake and exposed in inhalation chambers. Expired breath was collected from the artificially ventilated animals. Blood was sampled serially via venous cannulas during and after exposure.

CHEMICAL ANALYSIS

Headspace analysis was performed by GC with ECD (electron capture detection) and by combined GUMS. A 0.5 g or 0.5 ml sample was placed into a 23 ml headspace vial. The headspace vapor was cryofocused at subambient temperatures and analyzed on a PoraPLOT Q capillary column.

Urine samples from exposed animals were analyzed for fluoride by specific ion electrode. Urine from rats exposed to Halon 1211 were also analyzed for bromide by derivatization of the urine with dimethylsulfate to form methylbromide (1). Methylbromide was measured via combined GC/MS with subambient cryofocusing.

Urine samples were analyzed for carboxylic acid metabolites by derivatization to form volatile methyl esters (1). Derivatization was performed using dimethylsulfate, and analysis was done by combined GC/MS with subambient cryofocusing.

RESULTS

HALON 1211

Sprague Dawley rats exposed to Halon 1211 were found to have urinary bromide levels higher than control animals ($p < 0.05$). Control rats excreted 67 ± 13 µg ($n=4$) of bromide in 24 h, while exposed rats excreted 110 ± 24 µg ($n=4$) in 24 h. Urinary bromide from Fischer-344 was elevated, but not statistically different from the controls ($0.05 < p < 0.10$). The results of the urinary bromide analysis is shown in Figure 3.

PERFLUOROHEXANE

Metabolites of perfluorohexane were not detected in tissue samples, expired breath, or in the urine of Sprague Dawley or Fischer-344 rats. Immediately after exposure parent material was observed in tissues but 24 h following exposure perfluorohexane was only in the fat.

HCFC-123

HCFC-133a (2-chloro-1,1,1-trifluoroethane) was detected in the liver of Sprague Dawley and Fischer-344 rats exposed to HCFC-123 by nose-only inhalation. The total-ion-chromatogram and the mass spectrum of HCFC-133a are shown in Figures 4 and 5, respectively. Trace amounts of 2-chloro-1,1-difluoroethylene (CDE) were found in the liver of Fischer-344 rats exposed to HCFC-123. The total-ion-chromatogram and mass spectrum of CDE are shown in Figures 6 and 7, respectively. The analysis of urine from Sprague-Dawley and Fischer-344 rats were found to contain trifluoroacetic acid (TFA). The total-ion-chromatogram and mass spectra of the volatile methyl ester of TFA are shown in Figures 8 and 9, respectively. In a 24 hour period 2050 ± 570 µg ($n=4$) of TFA were excreted in the urine.

Gas uptake studies done at HCFC-123 concentrations of 0.01%, 0.1%, and 1% indicated that the metabolism of HCFC-123 occurs via a saturable pathway(s). These data, along with experimentally obtained partition coefficients were used to construct a preliminary physiologically based pharmacokinetic (PBPK) model describing the metabolism of HCFC-123.

This model indicated that the rate of HCFC-123 metabolism saturates at about 0.1% (1000 ppm) as shown in Figure 10.

The analysis of HCFC-123 in blood was performed during and after 4 h exposures to HCFC-123 at levels of 0.01%, 0.1%. and 1%. The results of these exposures (shown in Figure 11) gave log-concentrations vs time curves of HCFC-123 which increased proportionally with the respective exposure levels. However, the blood levels of TFA obtained during the same exposure experiment gave log-concentrations vs time curves indicative of saturated metabolism. As shown in Figure 12, the curve for the 0.01% exposure gave a steady state TFA concentration of about 30,000 µg/L while the 0.1% and 1% exposure groups gave nearly identical steady state TFA concentrations of about 110,000 µg/mL.

Preliminary data for the volatile metabolite, HCFC-133a were collected from expired breath of rats exposed to 0.01%, 0.1%. and 1% HCFC-123 for 2 h (Figure 13). The expired breath levels of HCFC-133a were lowest for 0.01% exposure group and similar in value for the 0.1% and 1% exposure groups.

CONCLUSIONS

The inhalation exposure studies conducted with perfluorohexane did not yield any indication of the metabolism of this compound. However, the investigation of Halon 1211 did reveal increased *urinary* bromide in urine of Sprague Dawley rats. The product of the debromination of Halon 1211 is chlorodifluoromethanewhich may be expired or oxidatively metabolized. **A** proposed metabolic pathway for Halon 1211 is shown in Figure 14.

HCFC-123 undergoes reductive dehalogenation to form HCFC-133a (2-chloro-1,1,1-trifluoroethane) and CDE **(2-chloro-1, l-difluoroethylene).A** proposed metabolic scheme for HCFC-123 is shown in Figure 15. The metabolite HCFC-133a has been shown to be toxic. Oral gavage studies done on Wistar derived male and female rats (300 mg/kg, 5 days/week for 1 yr) found uterine adenocarcinomas, and benign interstitial cell neoplasms of the testis (2). HCFC-123 also undergoes oxidative metabolism. **As** previously reported HCFC-123 (3) is oxidized to **TFA,** which was found in urine.

The preliminary **PBPK** modeling of HCFC-123 metabolism indicated that this compound has a saturable rate of metabolism which occurs at about 0.1% (1000 ppm). Evidence for this was seen in the concentrations of **TFA** in blood, and HCFC-133a in expired breath. Both of these metabolites had low concentrations at an exposure levels of 0.01%, which is below saturation, and increased concentrations at an exposure level of 0.1% which is near saturation. However, increasing the exposure level to 1.0% did not appreciably increase the concentrations of **TFA** in blood or HCFC-133a in expired breath. These data indicate that the metabolism of HCFC-123 becomes saturated at inhalation exposure levels of about 0.1% (1000 ppm). Inhalation exposure to higher levels of HCFC-123 will not result in an increased rate of metabolism.

REFERENCES

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2. "Hydrofluorocarbons and hydrochlorofluorocarbons Hazard Assessment," Office of Toxic Substances, Health and Environmental Review Division, U.S. Environmental Protection Agency, Washington, D.C. 20460, November 16,1990.

3. Harris, J.W., L.R. Pohl, J.L. Martin, and, M.W. Anders, "Tissue acylation by the chlorofluorocarbon substitute HCFC-123," The Toxicologist, 11(1):Abstract 1269, pg. 349 (1991).

Legends For Figures

Figure 1. Chemical structures of HCFC-123, Halon 1211 and Perfluorohexane.

Figure 2. Exposure methodology for metabolism study.

Figure 3. Urinary bromide data from 2 h, 1% Halon 1211 exposure.

Figure 4. Total-ion chromatogram of a liver headspace sample obtained from a F-344 rat exposed to HCFC-123. HCFC-133a has a retention time of 13.2 min and HCFC-123 has a retention time of 18.6 min.

Figure 5. Mass spectrum of HCFC-133a obtained from a liver headspace sample of a F-344 rat exposed to HCFC-123.

Figure 6. Total-ion chromatogram of a liver headspace sample obtained from a F-344 rat exposed to HCFC-123. 2-Chloro-1,1-difluoroethylene (CDE) has a retention time of 10.3 min; also shown is HCFC-133a with a retention time of 13.2 min.

Figure 7. Mass spectrum of 2-chloro-1,1-difluoroethylene (CDE) obtained from a liver headspace sample of a F-344 rat exposed to HCFC-123.

Figure 8. Total-ion chromatogram of a dimethylsulfate-derivatized urine sample obtained from a F-344 rat exposed to HCFC-123. The methyl ester of TFA has a retention time of 15.9 min.

Figure 9. Mass spectrum of the methyl ester of TFA from a dimethylsulfate-derivatized urine sample obtained from a F-344 rat exposed to HCFC-123.

Figure 10. Concentration-response curve from PBPK model of HCFC-123 metabolism. The curve shows the metabolic rate as a function of concentration at exposure levels of 100 ppm (0.01%), 1000 ppm (0.1%), and 10,000 ppm (1%) HCFC-123.

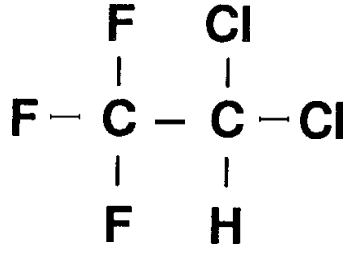
Figure 11. Log-concentration of HCFC-123 in the blood of rats after a 4 h exposure to HCFC-123 at exposure levels of 0.01%, 0.1%, and 1%.

Figure 12. Log-concentration of Trifluoroacetic acid in the blood of rats after a 4 h exposure to HCFC-123 at exposure levels of 0.01%, 0.1%, and 1%.

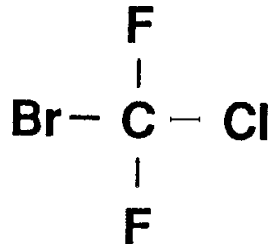
Figure 13. Concentration of HCFC-133a in expired breath of rats exposed to HCFC-123 for two hours. Measurements were made both during and post exposure.

Figure 14. Proposed metabolic pathway for Halon 1211. Reductive metabolism by cytochrome P-450 yields bromide and chlorodifluoromethane (Freon 22). The oxidation of Freon 22 may yield formylfluoride which would be rapidly hydrolyzed to HF and CO_2.

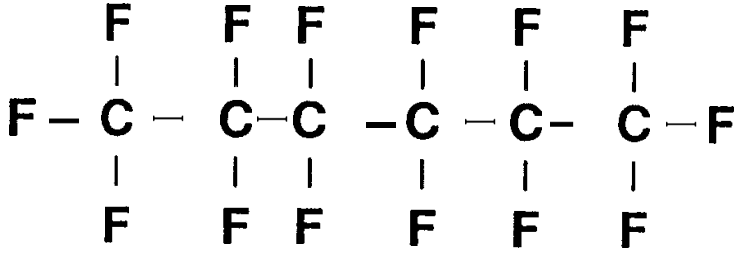
Figure 15. Proposed metabolic pathway for HCFC-123 showing reductive metabolism to HCFC-133a and CDE, and oxidative metabolism to TFA.



HCFC-123



Halon 1211



Perfluorohexane

Figure 1

Exposure Conditions

- 2 Hour 1% Nose-only Exposure
- 8 Test Animals
- 2 Controls (ambient air)

Sacrifice

- 4 Test Animals
- 1 Control
- Collect Tissue Samples

24 Hour Metabolism

- 4 Test Animals
- 1 Control
- Collect 24 Hour Urine and Feces
- Sacrifice Animals

Figure 2

Urinary Bromide (Br^-)

Halon 1211:	2h 1% Exposure
Sprague-Dawley Controls:	27 ± 13 µg/24 h
Sprague-Dawley Exposed:	150 ± 24 µg/24 h
Fischer 344 Controls:	73 ± 21 µg/24 h
Fischer 344 Exposed:	106 ± 22 µg/24 h

Figure 3

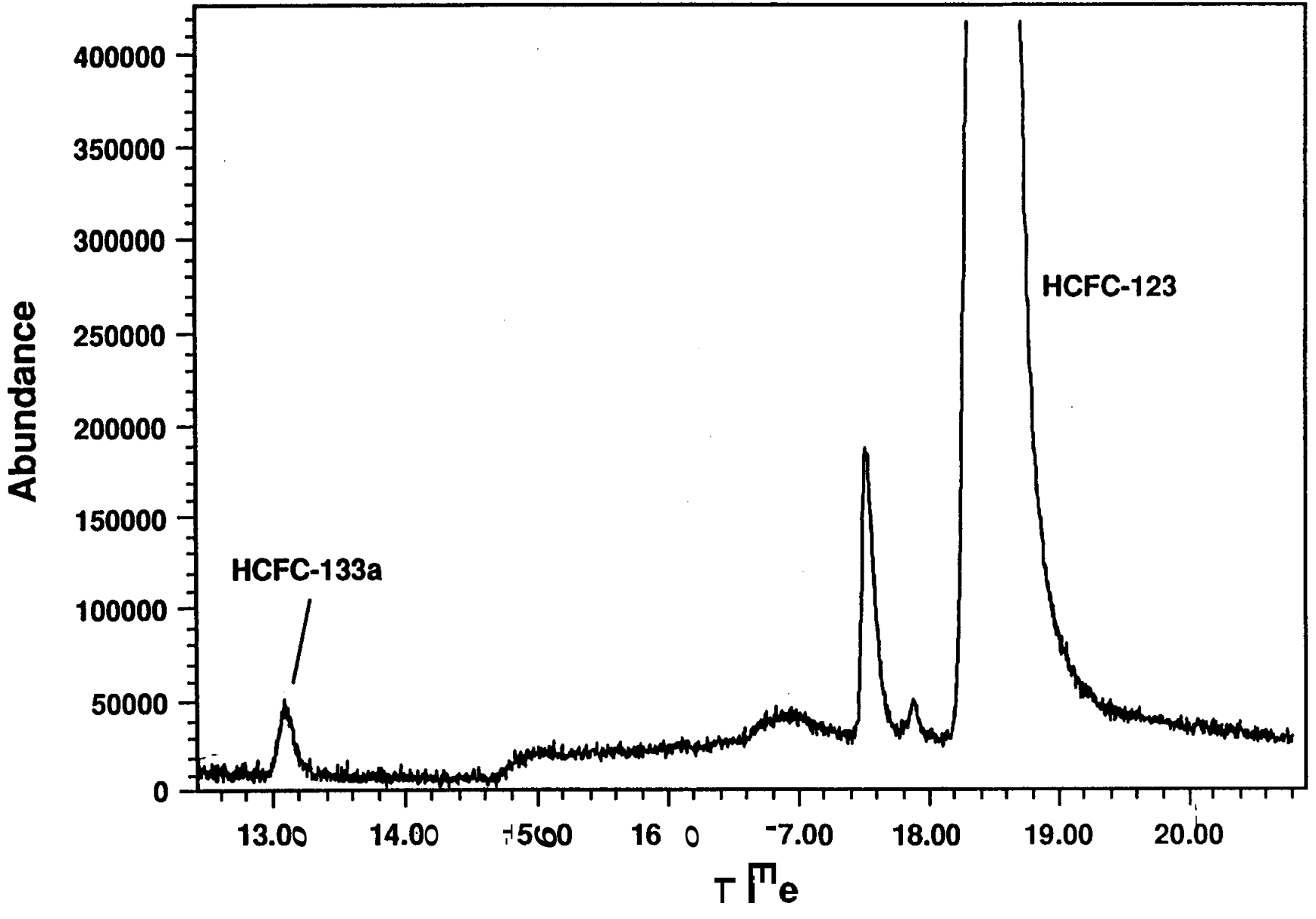


Figure 4

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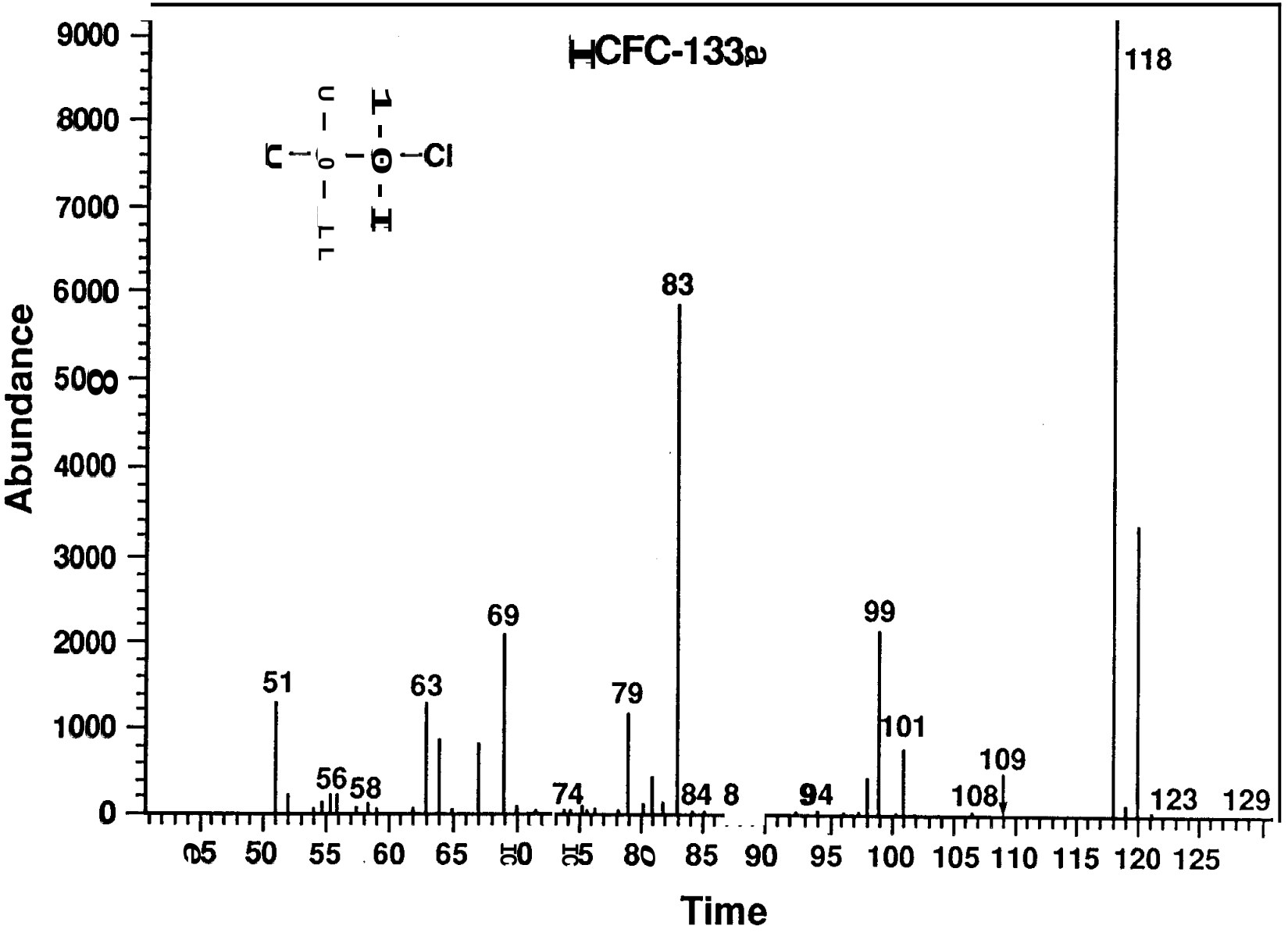
HCFC-133a
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5000
4000
3000
2000
1000
0
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69
99
51
63
79
101
109
56
58
74
84
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Figure 5

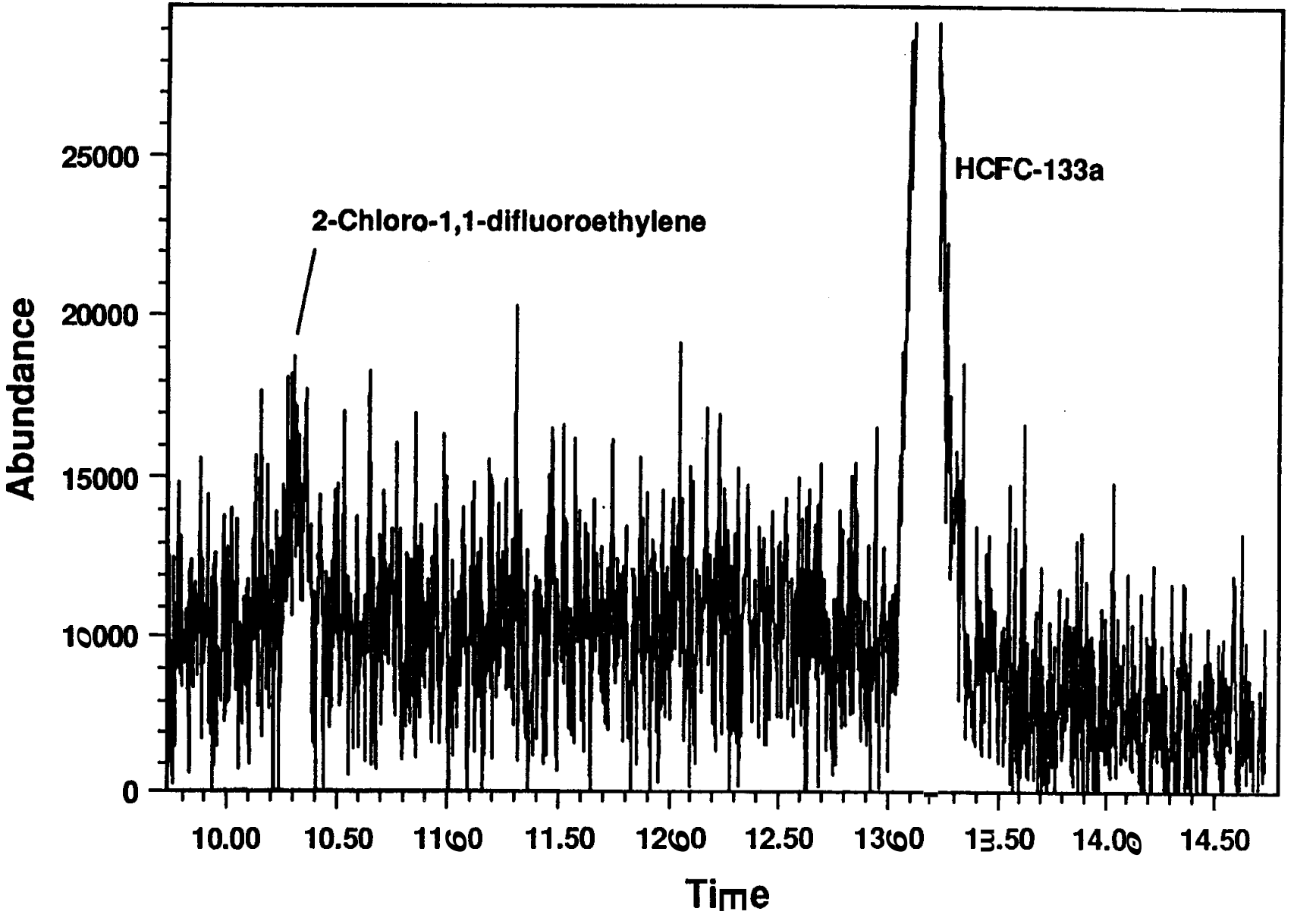

Figure 4

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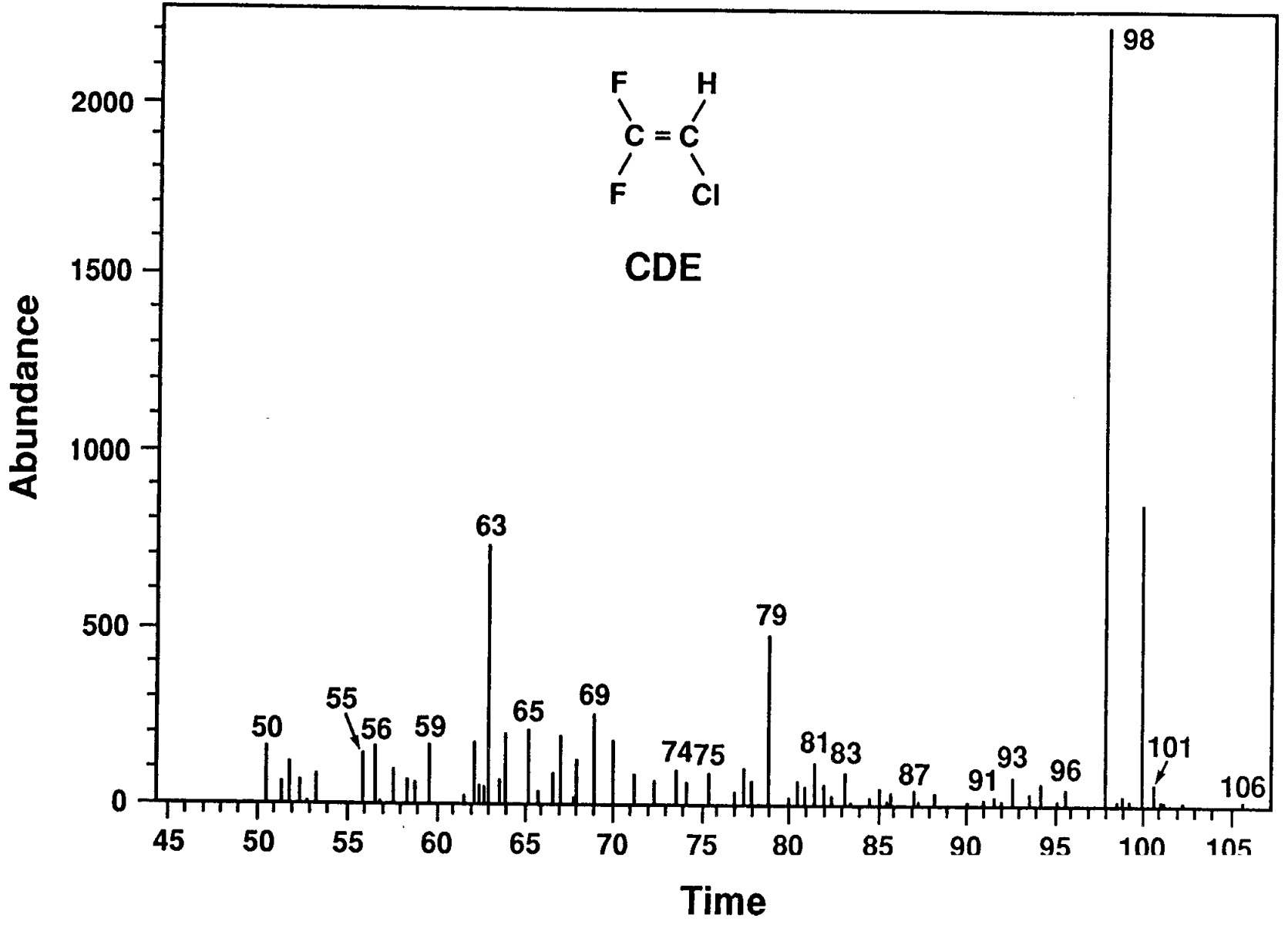
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Abundance
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Time

Figure 7

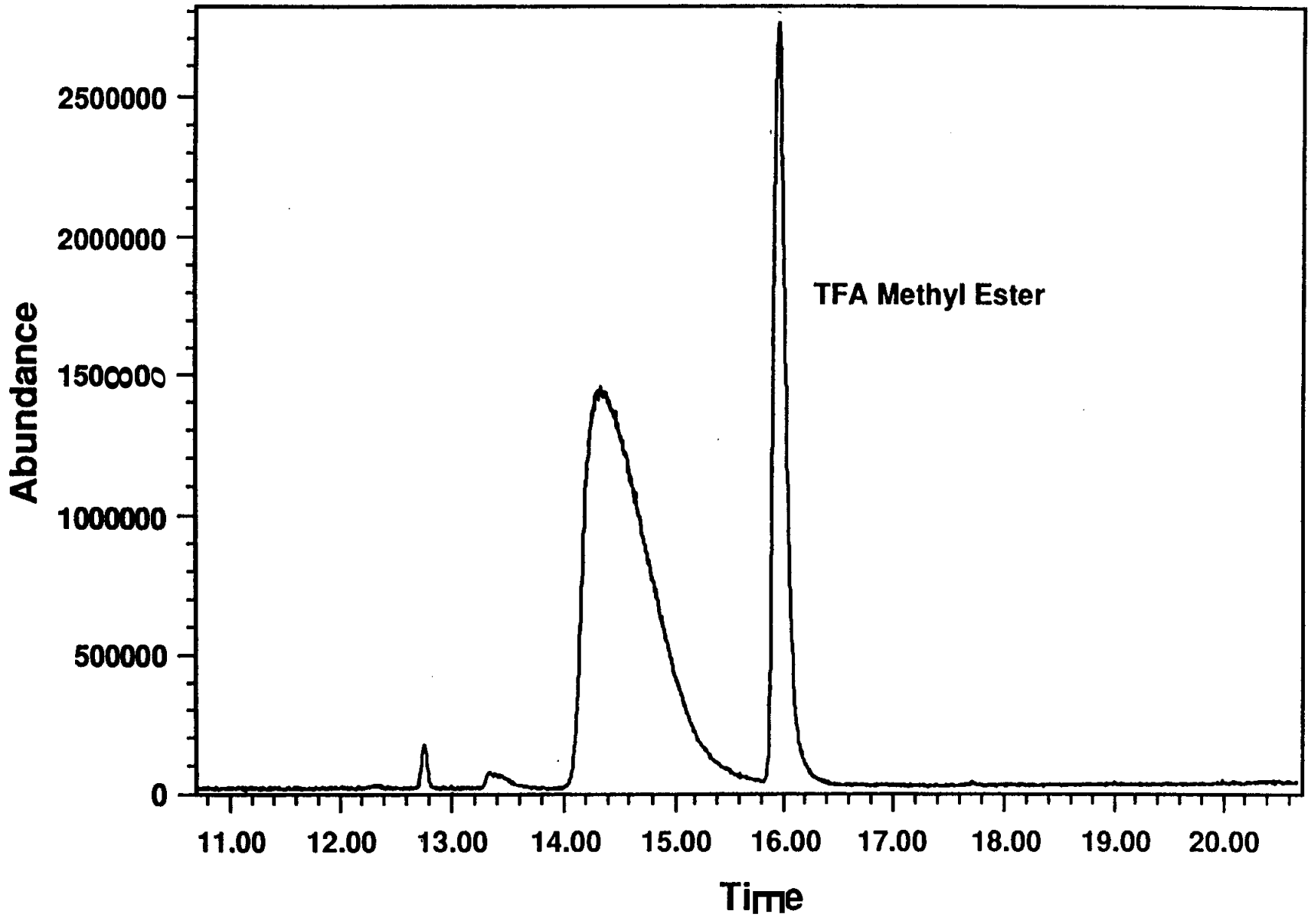
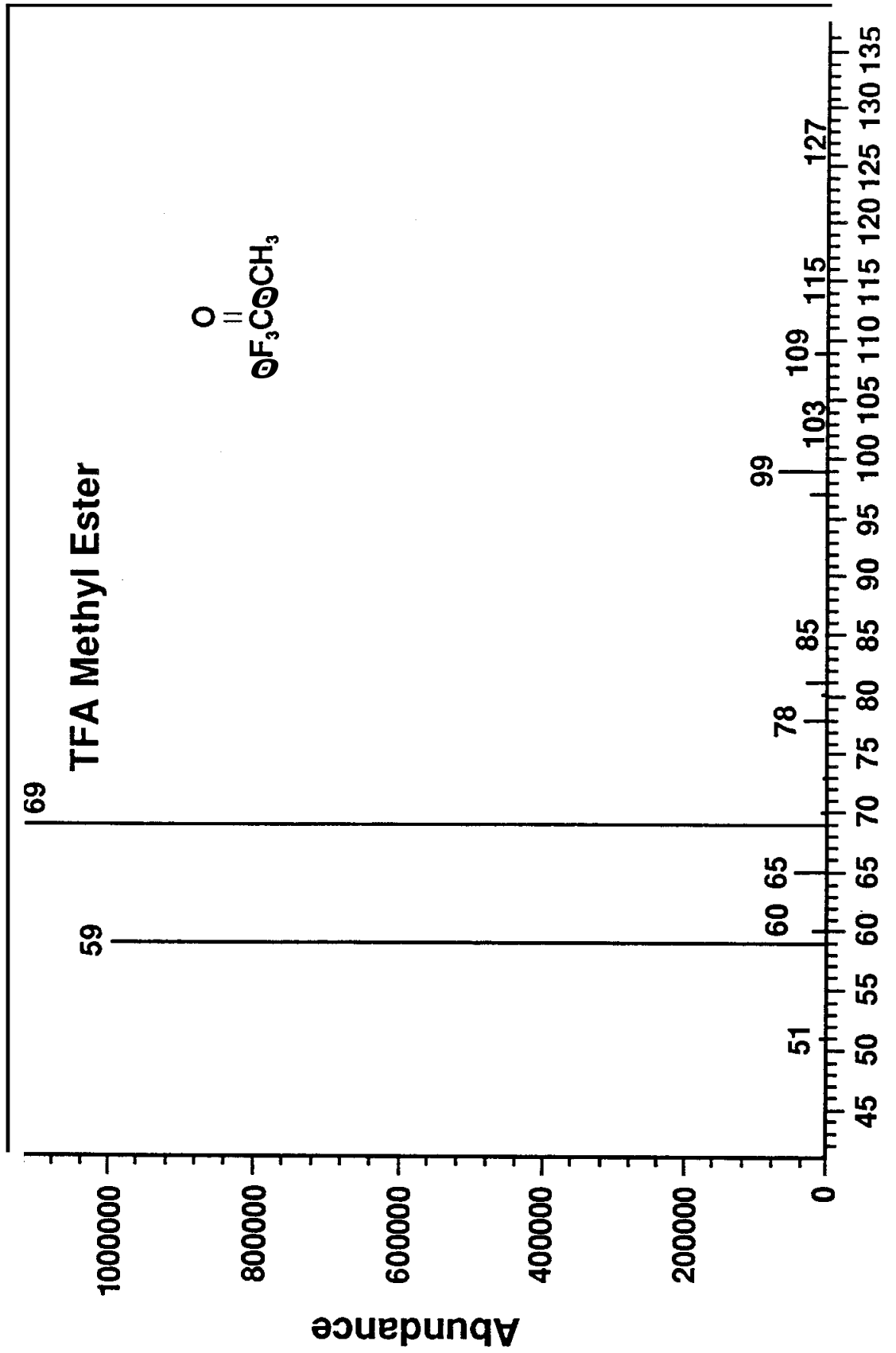


Figure 8

W. Brashear Fig. 6 4-22-50/92 ᑐ


TFA Methyl Ester
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60 65
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103
109
115
127
Abundance
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Time

Figure 9

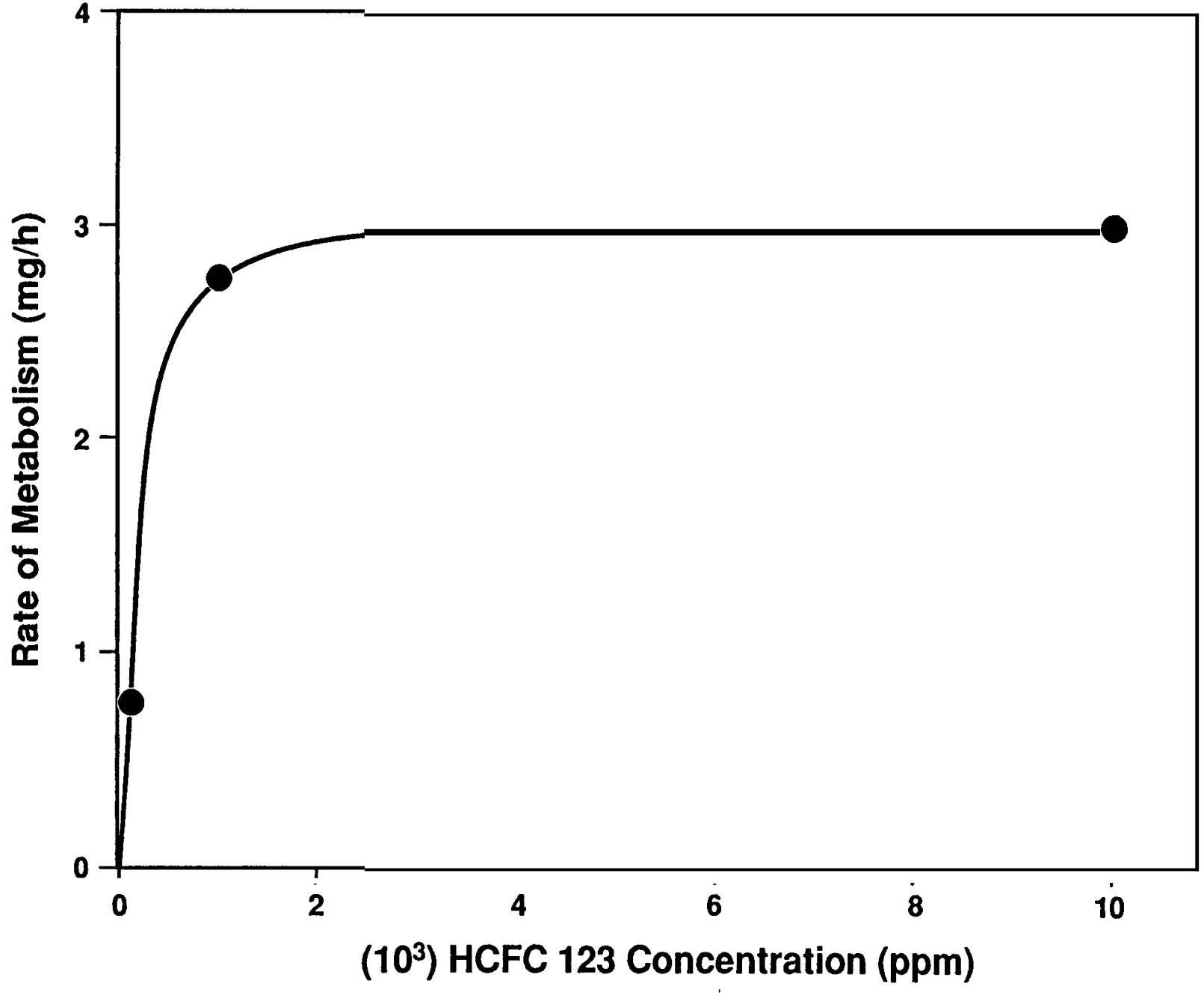

Figure 10

W. Brashear Fig. 8 4-22-50/92 CJ

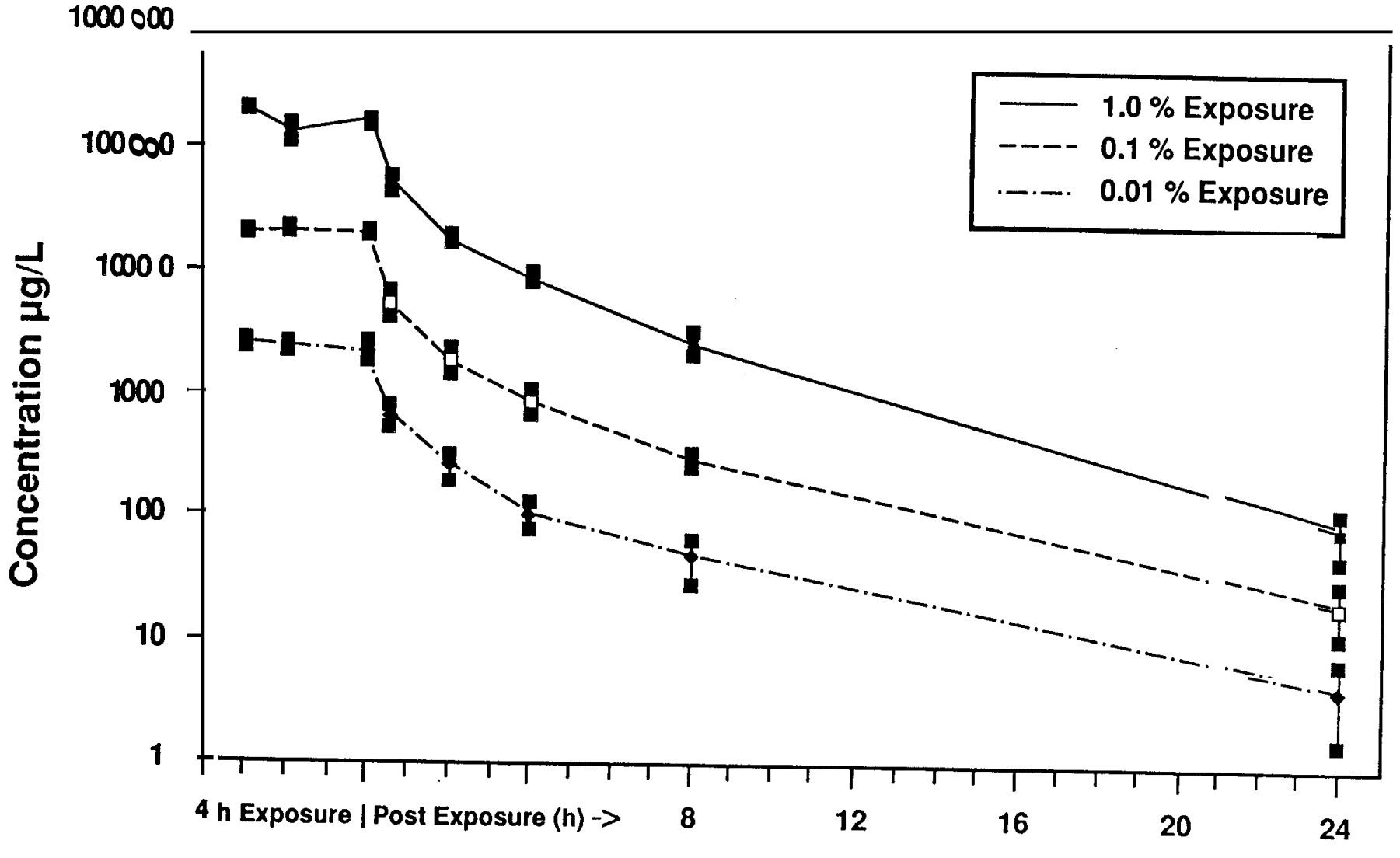
HCFC-123 Blood Concentrations
1000 000
100 000
1000 0
1000
100
10
1
Concentration µg/L
1.0 % Exposure
0.1 % Exposure
0.01 % Exposure
4 h Exposure | Post Exposure (h) ->
8
12
16
20
24

Figure 11

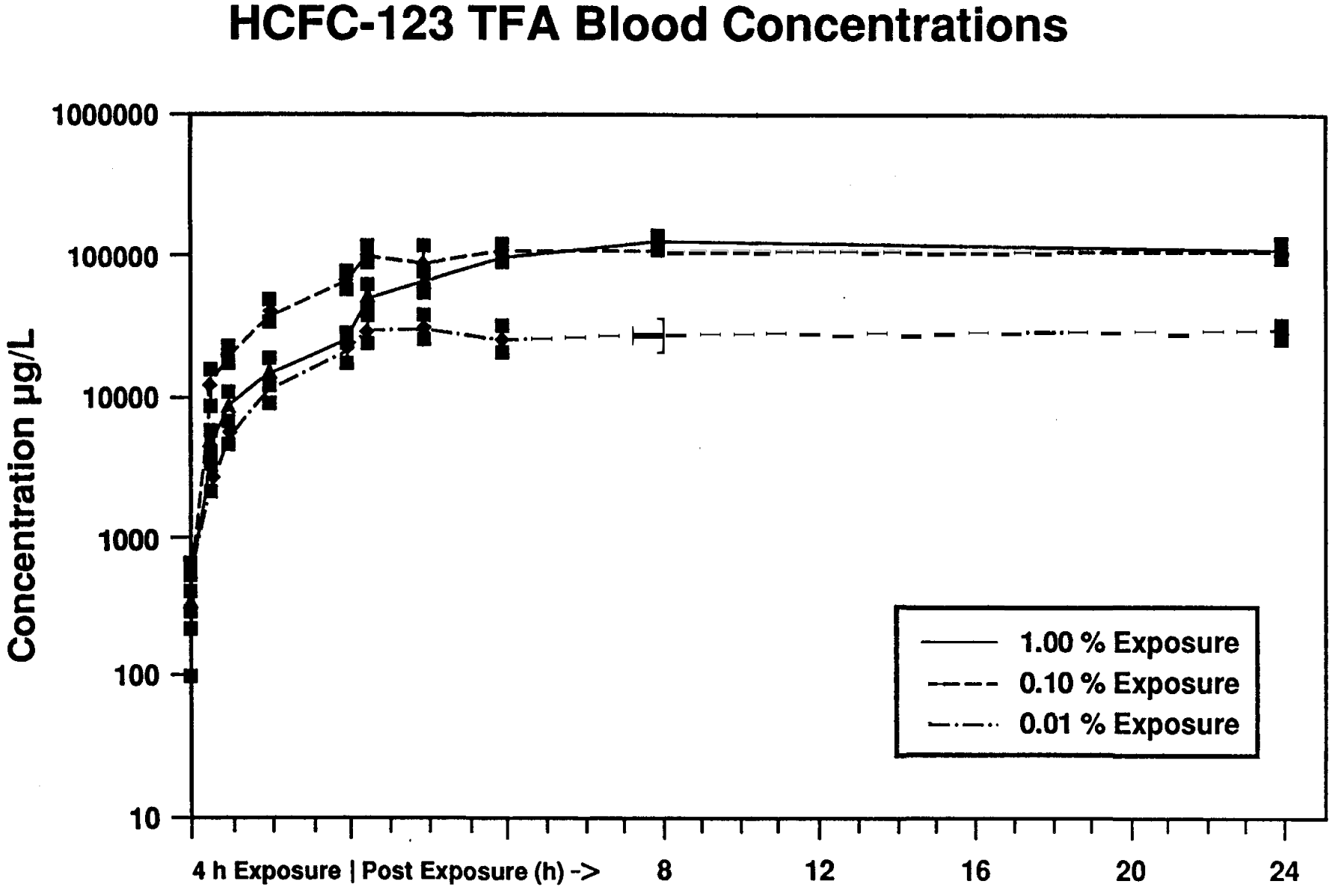

Figure 12

W. Brashear Fig 18 4-22-50/92 ω

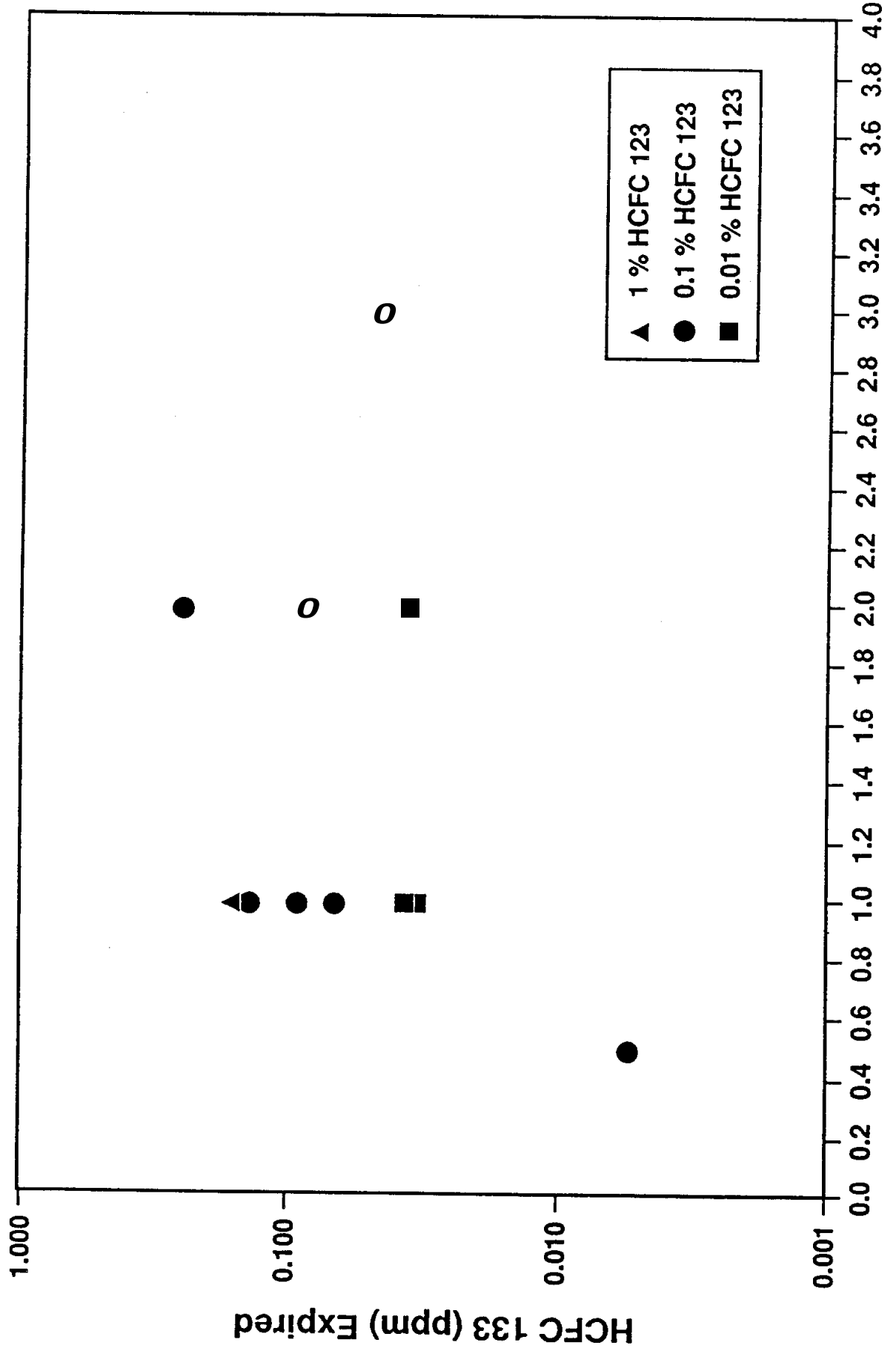

Figure 13

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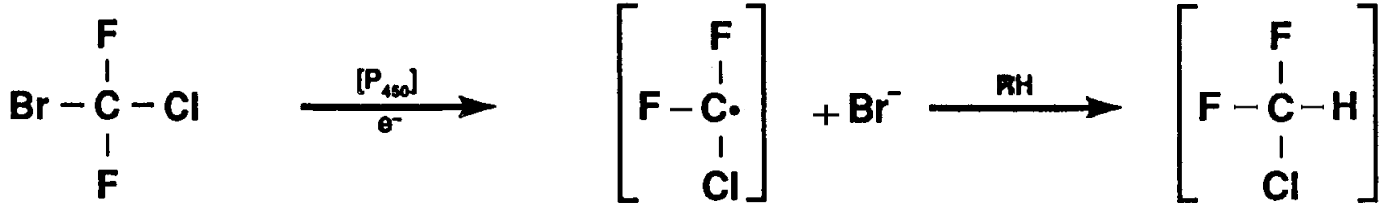

Halon 1211

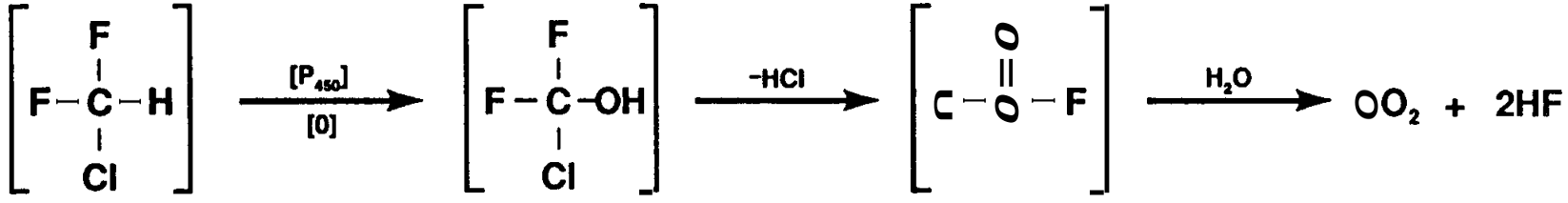

Figure 14

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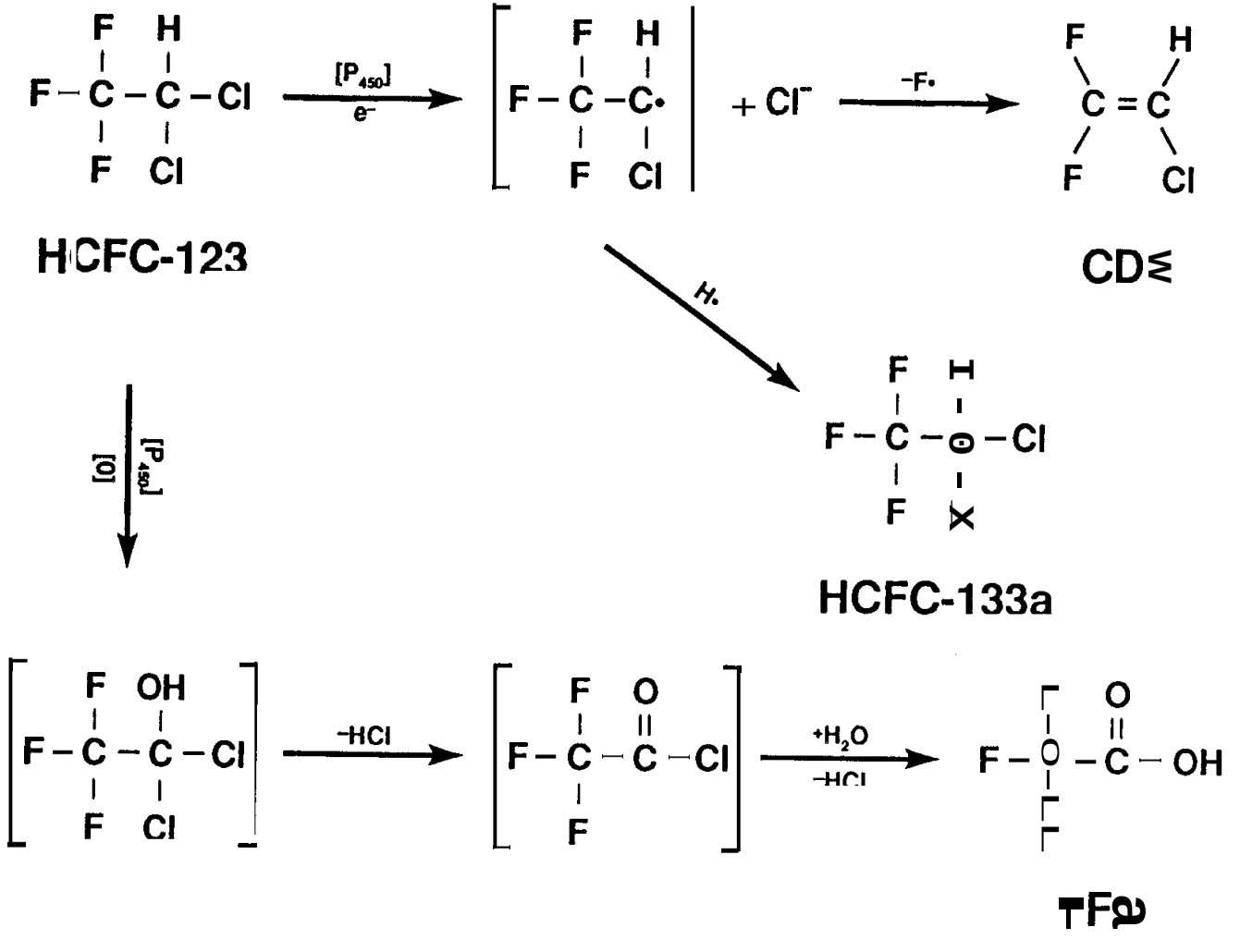

Figure 15

W. Brashear Fig.11 4-22-50/92 ω