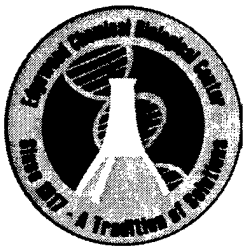

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NMR METHOD FOR THE QUANTITATIVE ANALYSIS OF VX HYDROLYSATES

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Disclaimer

The findings in this report are not to be construed as an official Department of the Army position unless so designated by other authorizing documents.

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14. ABSTRACT
This procedure is based on previously published ERDEC-TR-449, Nuclear Magnetic Resonance (NMR) Analysis of Chemical Agents and Reaction Masses Produced by their Chemical Neutralization and unpublished work entitled NMR Method for the Quantitative Purity Analysis of Feedstock Samples. This procedure uses an internal standard to establish an absolute weight percent for the analytes in question. Identifying the structures of other components in the mixture is not necessary. The weight percent calculations are not negatively affected by the presence of undetectable components (e.g., elemental sulfur, inorganic salts, etc.) in the sample.

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PREFACE

This work described in this report was authorized under Project No. 62262255200. The work was started in October 2001 and completed in September 2002.

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NMR METHOD FOR THE QUANTITATIVE ANALYSIS OF VX HYDROLYSATES

1. INTRODUCTION

1.1 Purpose.

This procedure is based on the previously published ERDEC-TR-449* and unpublished data.** This procedure uses an internal standard to establish an absolute weight percent for the analytes (Table 1) in question. Identifying the structures of other components in the mixture is not necessary. The weight percent calculations are not negatively affected by the presence of undetectable components (e.g., elemental sulfur, inorganic salts, etc.) in the sample.

Table 1. Analyte List

DCH urea, other CDT products
DIPA
EMPA
EMPSH
EtOH
MPA
Other RS-compounds
RSCCSH
RSH
RSR
RSSR
P-H
Phosphates (δ 0-19)
Other acids/esters (δ = 19-39),
RP(O)(SR), R2P(S) (δ = 39-69)
Other P=S (δ = 69-100)
CH3P(O)(OH)H
31P compounds (Total)
RSEt
Other 31P
EA-2192

* Brickhouse, M.D.; Rees, M.S.; O'Connor, R.J.; Durst, H.D. *Nuclear Magnetic Resonance (NMR) Analysis of Chemical Agents and Reaction Masses Produced by their Chemical Neutralization;* ERDEC-TR-449; U.S. Army Edgewood Research, Development and Engineering Center: Aberdeen Proving Ground, MD, 1997; UNCLASSIFIED Report (AD-A339 308).

** O'Connor, R.J.; Brickhouse, M.D.; McGarvey, D.; Durst, H.D.; Creasy, W.R.; Ruth, J.L. *NMR Method for the Quantitative Purity Analysis of Feedstock Samples*; ECBC-TR-253; U.S. Army Edgewood Chemical Biological Center: Aberdeen Proving Ground, MD, 2002; UNCLASSIFIED Report (AD-A406 815).

1.2 Analyte Concentration Range.

Analytes can be quantified at any concentration level from the detection limit to neat material. The following detection limit study was performed using phosphorus NMR on the Bruker AC-300 (Table 2). Detection limits for other nuclei and other instruments were estimated using the knowledge of the operator. Detection limits will depend on the matrix and interferences. If detection limits are required for a particular measurement, they should be determined for the particular matrix, analyte, and instrument and given in the analysis report.

Table 2. ACWA P&A Results for 100 ppm DIMP Standard in D_2O on AC-300 Results for Standard in DI Water by ^{31}P NMR, 8 Runs on Each of Two Days

Sample No. NB130P24B
Integration from NUTS, IS=HMPA, compound is DIMP, EM=1, manual phase

File	Area for Internal Standard (0.26 for P)	Area of compound peak	Wt. %	µg/g sample (ppm)
AR090F.114	100000	83.47	0.00719	71.95
AR091F.114	99175	95.02	0.00826	82.58
AR092F.114	100354	100.33	0.00862	86.17
AR093F.114	100714	146.86	0.01257	125.69
AR094F.114	100616	150.1	0.01286	128.59
AR095F.114	101055	142.02	0.01211	121.14
AR096F.114	99364	92.78	0.00805	80.48
AR097F.114	99201	92.35	0.00802	80.24
AR090F.116	96716	69.87	0.00623	62.27
AR091F.116	98331	146.85	0.01287	128.73
AR092F.116	99423	147.28	0.01277	127.68
AR093F.116	100302	150.89	0.01297	129.67
AR094F.116	100915	138.24	0.01181	118.08
AR095F.116	98958	101.63	0.00885	88.52
AR096F.116	98634	84.36	0.00737	73.72
AR097F.116	98668	72.6	0.00634	63.42
Average Found				98.06
Concentration (ppm) Standard deviation				26.20
MDL (ppm)				68.18
Sample concentration				99.48
Recovery %				98.57%

1.3 Sample Matrices and Interferences.

This method can be performed on liquid matrices that solubilize the internal standard. Generally, either TEP (triethylphosphate) or HMPA (hexamethylphosphoramide) have been soluble in each AWCA matrix. Other internal standards may be used, as long as they are stable and the signals from the standard do not interfere with other peaks in the spectra. For basic hydrolysate matrices, TEP is the preferred internal standard. They must also be commercially available with a high purity.

Paramagnetic metals such as iron and chromium at fairly low concentrations will broaden the peaks and either increase the error of the analysis, or invalidate it entirely. Ongoing research on the use of Tiron as an iron chelator and spin-relaxation agent may partially solve this problem in the future.

1.4 Throughput.

Sample throughput depends on the nucleus that is being used for the analysis, and the sensitivity required. A typical experiment may allow three samples to be analyzed in an 8-hr day.

2. RISK AND SAFETY ASSESSMENT

All analyses should be performed in accordance with all appropriate Federal, State and local laws, as well as Army Regulations. Samples containing chemical agent should be handled in accordance with AR 50-6 and all other applicable regulations. Exposure to chemical agents or other super toxic materials may result in injury or death.

3. SCIENTIFIC BASIS

NMR spectroscopy has been a proven method for the identification and quantification of chemical materials for many years.*

4. TRAINING

Operators should have a Master's degree in chemistry, or the equivalent in work experience. Specific training in the use of the instrument, as well as in the handling of hazardous materials, should be obtained.

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* Silverstein, R.M.; Bassler, G.C.; Morrill, T.C. *Spectrometric Identification of Organic Compounds*; 5th ed., John Wiley and Sons, Inc.: New York, 1991.

5. APPARATUS

5.1 Instrumentation.

Any make or model of NMR spectrometer may be used. Sensitivity and selectivity will depend on the field strength, type of probe, and processing software.

5.2 Glassware, Miscellaneous Equipment and Supplies.

NMR sample tubes such as the 5 mm Wilmad 507-PP or the 535-PP may be used.

5.3 Chemicals.

Deuterated solvents and internal standards may be purchased from commercial vendors, such as the Aldrich Chemical Corporation. Deuterated solvents may be kept for years if properly stored in a sealed container. Internal standards should be kept in sealed containers and kept dry. Hydrolysis of the internal standards would create additional peaks in the spectra and affect the quantification of the other analytes. A certificate of analysis should be obtained with each bottle of standard material.

6. PROCEDURE

6.1 Sample Preparation.

This procedure should be performed under appropriate engineering controls, in accordance with all appropriate surety and safety regulations.

a. If the sample contains two layers, each should be analyzed separately.

b. Tare a screw-cap vial and cap. Dispense ~0.1mL of neat internal standard (usually TEP for acidic samples, TEP or HMPA for neutral or basic samples) of known purity into the vial and replace the cap. The exact weight is determined using an analytical balance capable of measuring to the hundredth of a milligram. Record the weight, and tare the balance.

c. Add 0.1-1.0mL of the neat sample into the vial. Replace the cap and record the weight. Add 0.1mL D_2O as a lock solvent for aqueous samples. Non-aqueous samples could be prepared using an organic solvent such as $CDCl_3$. Samples that are reactive or insoluble in common solvents may be analyzed without the use of a lock signal, or by the use of a coaxial insert.

d. Mix the sample vial on a vortexing apparatus to assure homogeneity.

6.2 Obtaining NMR Spectra.

Operating parameters vary according to the sample. The sample spectra in the appendix show example parameters that may be used. The important parameters to consider include the following:

- *delay time* should be 5-10 times the T1 value for maximum quantitation accuracy, but shorter delay times can be used to improve signal to noise ratios and sensitivity with less accurate quantitation;

- *line broadening* should be large enough to give acceptable line shapes and baseline flatness;

- *number of scans* is determined by the signal to noise ratio that is required and limits on analysis time;

- *number of data points* is determined by the required spectral resolution to resolve interferences;

- *pulse width PW90 and amplifier gain* are adjusted to give the largest signal without saturating the detector electronics;

- *spectral width* must be wide enough so that peaks of interest are not near the edge of the spectrum.

a. Place the NMR tube into the spinner, using vendor supplied depth gauge to orient sample at maximum sensitivity position to the coils inside the probe. Lower the sample into the magnet, lock onto the deuterium signal and shim to maximize lock signal. If no deuterated solvent is used, the operator may shim on the FID, or by observing the processed spectrum.

b. Tune probe for optimal frequency and impedance match for the sample.

c. To correctly set up the instrument parameters, it is necessary determine the PW90 for each nucleus, and the T1 values, unless those values are known from similar samples run previously.

d. Enter sample name and information into NMR operating system and acquire data. If necessary, setup multiple samples on the autosampler with careful attention to placing samples in the correct positions. Acquire data for all samples by ^{1}H (detection limit 20-150 ppm). To save instrument time, if no analytes are detected, do not run ^{13}C. At the discretion of the analyst, ^{13}C NMR may be used as an additional tool for the identification and quantification of some analytes. It is particularly useful when iron contamination or other factors broaden the proton peaks to the point where the spectrum is not interpretable. ^{31}P NMR spectra should be obtained for all samples.

6.3 Spectral Data Analysis.

Examples of processed data are provided in Figures 1-4.

a. Apply appropriate window functions such as exponential multiplication to enhance signal-to-noise (line broadening for ^{31}P and ^{13}C should generally be 1-5 Hz, and 0.5 – 1 for ^{1}H).

b. Fourier transform the resulting data to convert from time to frequency and produce the NMR spectrum.

c. Correct the baseline and phase all peaks in the spectrum.

d. Reference the chemical shift against the internal standard, if needed. (TEP should have a shift of 0.01 ppm for ^{31}P NMR).

e. Integrate all peaks in the spectrum to obtain areas.

7. CALCULATION, CALIBRATION, AND DOCUMENTATION

The NMR quality control standard (ethyl benzene or triphenylphosphate standards from Bruker Instruments) is analyzed to assure that the spectrometer meets signal to noise specifications and chemical shift criteria defined for the instrument.

The weight percent of each analyte for the sample can be calculated with the following equation when an internal standard (IS) is present:

$$\frac{\text{Area under analyte Peak}}{\text{Area under IS Peak}} \times \frac{\text{Molecular Weight of analyte}}{\text{Molecular Weight of IS}} \times \frac{\text{Weight of IS}}{\text{Weight of analyte}} \times 100\% = \text{Wt. \% Analyte}$$

If analytical precision is to be reported, from at least seven replicates calculate the mean and standard deviation. The mean value ± 2 standard deviations will provide a 95% confidence range. Generally, time does not allow for the acquisition of 7 spectra, and a single spectrum is used.

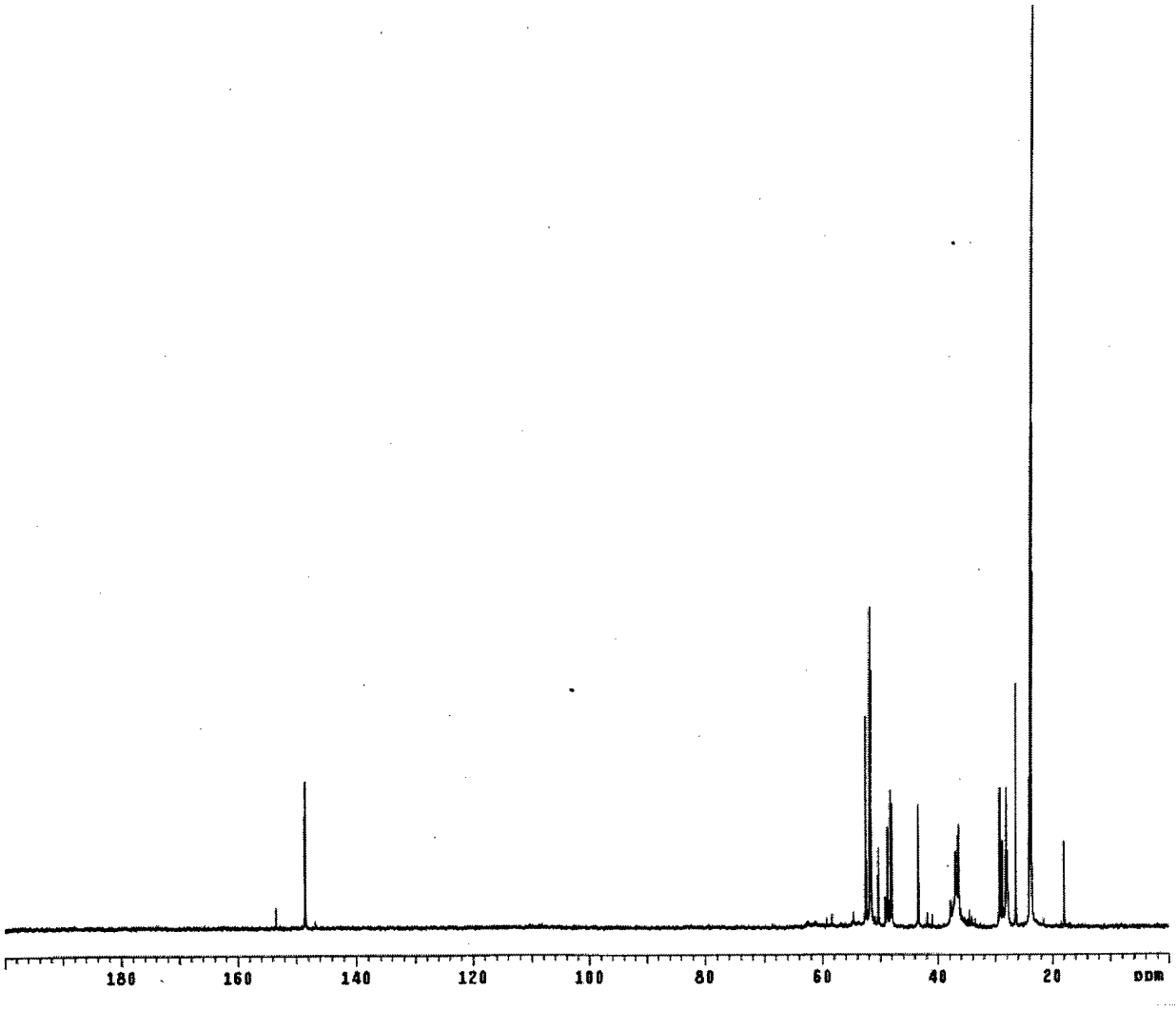

Figure 1. ^{13}C Spectrum of the Organic Layer of a VX Hydrolysate

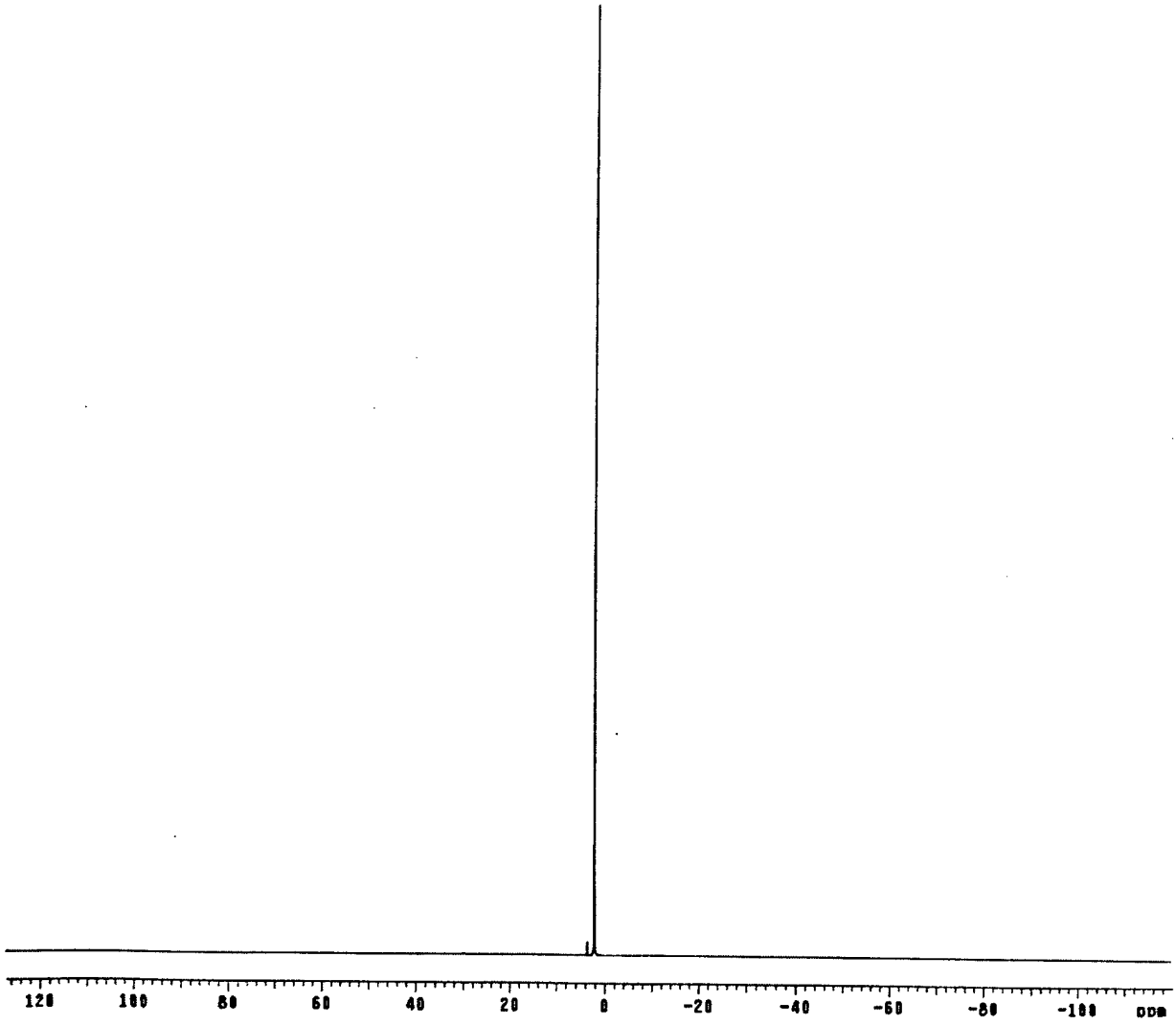

Figure 2. ^{31}P Spectrum of the Organic Layer of a VX Hydrolysate

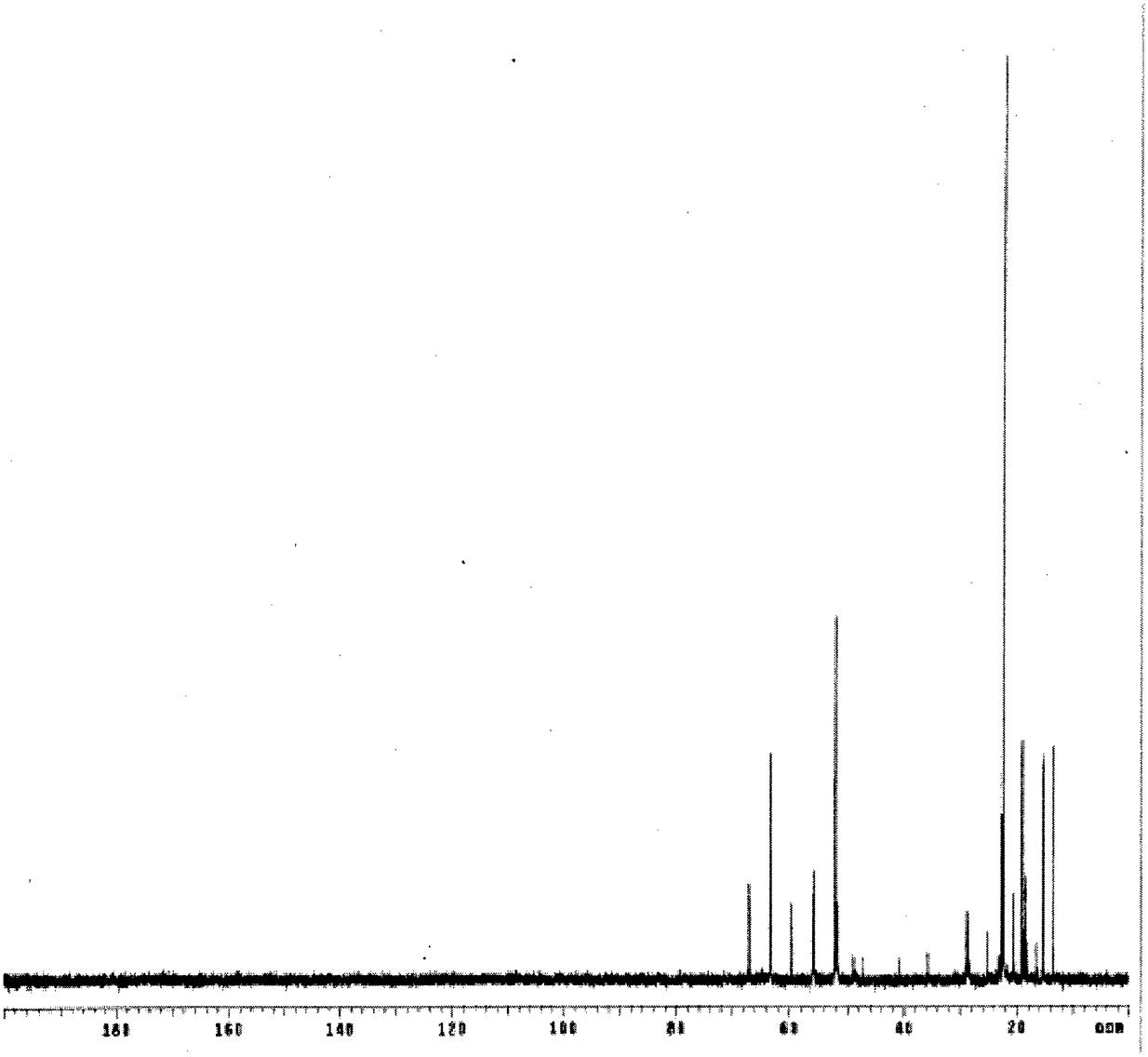

Figure 3. ^{13}C Spectrum of the Aqueous Layer of a VX Hydrolysate

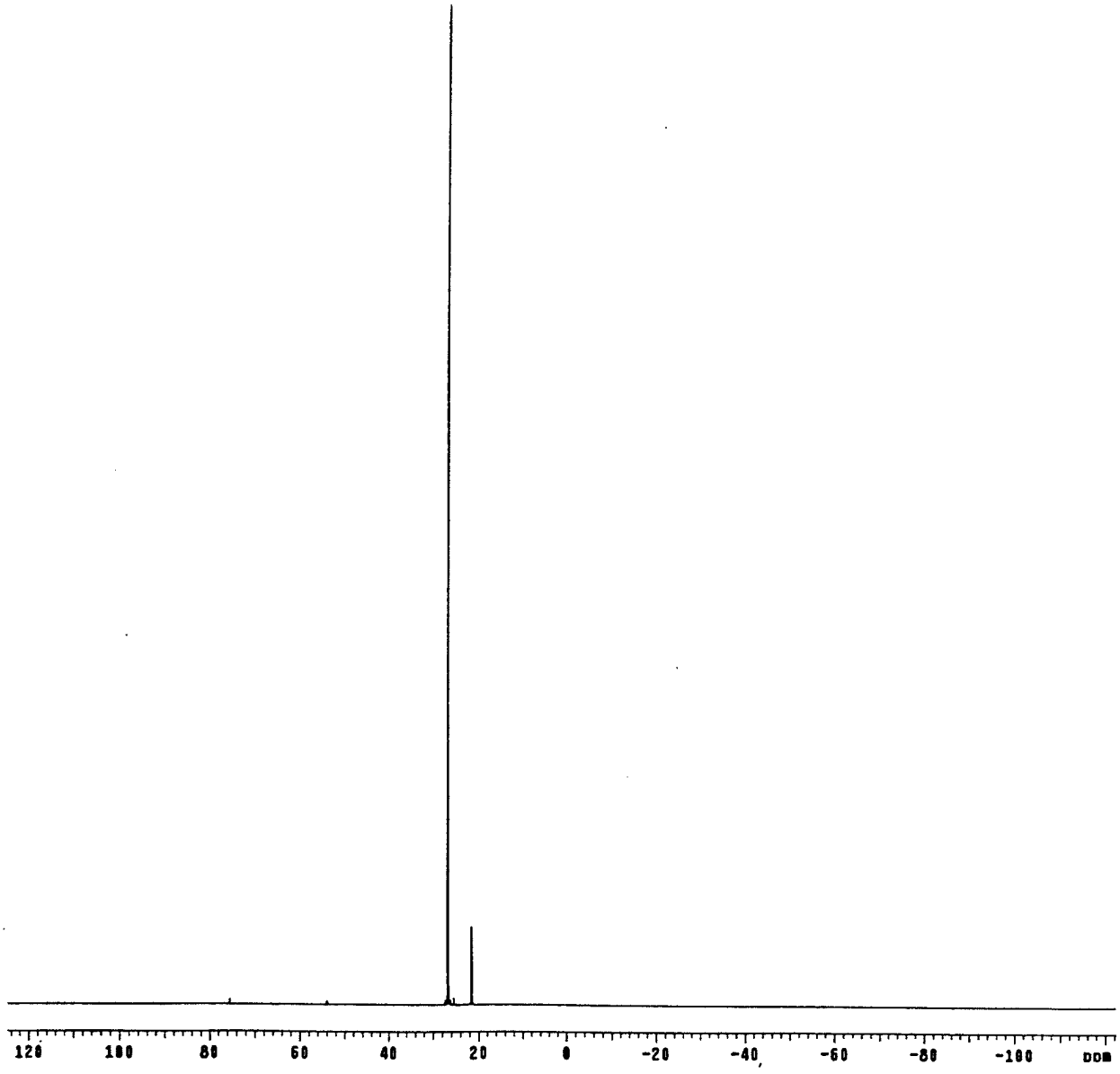

Figure 4. ^{31}P Spectrum of the Aqueous Layer of a VX Hydrolysate