CONTAMINANT SURVEY
OF THE
ANAHUAC NATIONAL WILDLIFE REFUGE

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Several people contributed to this monitoring survey of the Anahuac National Wildlife Refuge. Russ Clapper (retired) and the present refuge manager, Domenick Ciccone, provided access to the refuge, their personnel, and equipment. Jim Neaville and Ed Jackson, from the refuge staff, provided historical information about the refuge and field assistance time to insure our success with the collection of samples. Denise Baker and Larry Gamble helped with field collections, sample shipment, and other logistical problems. Charles Sanchez, Regional Contaminant Coordinator, helped with securing funds for analysis and getting the samples analyzed.

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ABSTRACT

Sediment and biotic samples were collected from four locations on the Anahuac National Wildlife Refuge for a contaminant survey. Contaminants examined in the analysis included organochlorine and organophosphate pesticides, heavy metals, and petroleum hydrocarbons. Analytical results indicate the refuge is, for the most part, not contaminated by these chemicals. Two localized areas have minor contaminant problems. Petroleum hydrocarbons contaminate the irrigation canal sediment near a diesel-powered lift pump and the bottom sediments of Jackson Ditch near a petroleum production area. Four heavy metals (chromium, copper, nickel, and silver) were also present at elevated levels in the sediment of Jackson Ditch, relative to other locations on the refuge.

INTRODUCTION

Anahuac National Wildlife Refuge was established in 1963 with the acquisition of 9,836 acres of marsh land bordering East Bay in Chambers County, Texas. In 1982, the refuge more than tripled in size with the purchase of a large segment of the Barrow Ranch, also in Chambers County (Fig. 1), and now totals 28560 acres. A variety of fish and wildlife species utilize this refuge from the shoreline on East Bay, through the brackish marshes, and into the rice farming areas on the northern section of the refuge.

Several bayous such as Oyster Bayou, Onion Bayou, East Bay Bayou, and Elm Bayou drain many square miles of farm and pasture land that border the refuge (Fig. 1). This surrounding land is farmed with rice, soybean, and some sorghum. The fallow rice fields are grazed by cattle, and there are scattered oil and gas production fields on all sides of the refuge. Contaminants that are associated with farming, ranching, and petroleum production have pathways to the refuge via the bayous. Many of the oil production facilities are permitted to discharge their brine (i.e., production water) into tidal canals and creeks both on, and near, the refuge. This brine water may have a salinity of 170 parts per thousand (ppt), whereas sea water is 35 ppt.

Rice farmers flood their fields during the growing season with fresh water delivered through a series of irrigation canals and drain the excess water through a network of drainage canals. This drainwater typically will enter into the nearest bayou and

See Table/Figure

FIGURE I. ANAHUAC NATIONAL WILDLIFE REFUGE, CHAMBERS COUNTY, TEXAS,

MAP

(SEE ORIGINAL)

flows through the refuge. Some of the drainwater is pumped into the irrigation canals used to flood the rice farming areas on the refuge. Rice farmers around the refuge, and those that lease the refuge lands, apply several herbicides and pesticides on these crops.

The refuge serves as a nesting area for the mottled duck (Anas fulvigula maculosa) and, during the winter months, as many as 20 species of ducks and geese may be found on the refuge. The rice farming area and the brackish marshes together provide wintering habitat for up to 50,000 geese. Drainage ditches, ponds, and bayous on the refuge provide habitat for marsh birds, fish-eating birds, migratory shore birds, and several species of commercially important finfish. Both sport fishing and waterfowl hunting are allowed on portions of the refuge.

Contaminants that may be impacting fish and wildlife species include: petroleum hydrocarbons such as aliphatic and polycyclic aromatic hydrocarbons; pesticides such as organochlorines and organophosphates; herbicides for aquatic vegetation control; and heavy metals. This study was designed to monitor sediments, aquatic invertebrates, fish, and birds for the presence of contaminants both on the refuge and in the bayous that deliver water to the refuge marshes.

METHODS

Sediment samples were collected from the bayous that border and drain into the refuge as access would permit. Sediment samples were also collected from waterways, rice farmland, and ponded water on the refuge. These samples were collected with a stainless steel petite ponar bottom sampler and stored on ice in one-liter chemically-cleaned glass jars until the sample could be frozen. All samples were frozen upon returning to the Clear Lake Field Office. Sampling stations are described in Table 1.

Blue crabs were collected from tidal canals using baited crab traps set for a 24-hour period. Several species of fish (carp, catfish, gar, shad) were collected using gill nets set in irrigation canals and bayous draining onto the refuge. Aquatic invertebrates (insect larvae, crayfish, shrimp) were collected using a small mesh 10-foot seine and a dip net in the rice drain ditches. Red-winged blackbirds were collected in the rice fields

using steel shot.

Samples for chemical analysis of the biota included whole body analysis of the invertebrates, livers from birds and fish, and viscera from the blue crabs. All samples were analyzed by contract laboratories through the Patuxent Analytical Control Facility (Laurel, MD). dive percent of the analyses were subsequently confirmed by gas chromatography/mass spectrometry (GC/MS). Table 2 is a list of all compounds considered in the analysis. Exact analytical extraction and determination procedures are available from the Patuxent Analytical Control Facility upon request and are not restated in this report. Brief descriptions are provided in Appendix I.

See Table/Figure

Table 1. Sampling stations and samples collected to monitor contaminants on the Anahuac National Wildlife Refuge, Texas.

(SEE ORIGINAL)

See Table/Figure

Table 2. Contaminants routinely analyzed in environmental samples by contract laboratories.

(SEE ORIGINAL)

RESULTS

A few pesticides and herbicides have been approved for use on the Anahuac National Wildlife Refuge for the rice farming areas. However, a survey of the local applicators produced a list of ten chemicals that were applied either on the refuge or on the adjacent fields (Table 3). In addition to this list, the herbicide 2,4-D was also sprayed in the drainage ditches on the refuge by Chambers County Drainage District. These 10 chemicals are all short lived in the environment, usually dredging below a detection limit within six weeks, thus they were not included in the 69 chemicals (Table 2) analyzed for in this contaminant survey.

HEAVY METALS

Sediment samples from four locations on the refuge (Table 4) did not reveal a serious problem with metal contamination. Eight of the 20 metals considered in the sediment analysis were below the detection level. Sediment samples from Jackson Ditch, a dredged canal used to drain the marshes into East Bay Bayou, had elevated

levels of chromium, copper, nickel, and strontium (Table 4). Jackson Ditch is now influenced by tidal action along the Gulf Intracoastal Waterway and may receives waste water from an oil and gas production area adjacent to the south end of the refuge. Strontium is one of the alkaline earth metals (Brady and Holum 1981) and is associated with saltwater, especially the produced water from petroleum production, Nickel and chromium are used in stainless steel production, and copper is a component of the antifouling paint used in shipping. These four metals detected at elevated levels are likely contaminants associated with the corrosion of metal equipment used along the Gulf Intracoastal Waterway by the shipping industry, and from the petroleum production industry.

There are very few published articles on strontium toxicity or levels of concern to fish and wildlife species. In contrast, chromium, copper, and nickel have extensive literature on their toxicity and accumulation potential in the environment (Phillips and Russo 1978). Chromium can exist in several valence states, but in aquatic environments it is usually found as trivalent or hexavalent chromium (U.S. EPA 1980a). Hexavalent chromium is a strong oxidizing agent and is very soluble in water which increases it's toxicity. Invertebrates are usually more sensitive to hexavalent chromium than are fish species and to

See Table/Figure

Table 3. Pesticides aerially applied on or near the Anahuas National Wildlife Refuge.

(SEE ORIGINAL)

See Table/Figure

Table 4. Mean heavy metal residues om twp sediment samples taken from four sites on the Anahuac National Wildlife Refuge.

protect these organisms the hexavalent chromium should not exceed 0.054 parts per million (ppm) in the water. The chromium level in the sediment from Jackson Ditch was 139.5 ppm, on a dry weight basis, which is 2,580 times the toxic dissolved level. This indicates that a potential chromium problem exists in Jackson Ditch.

Copper was elevated in the sediment samples from Jackson Ditch (Table 4). The criterion to protect saltwater aquatic life for copper is 0.04ppm as a 24-hour average, or not to exceed 0.023 ppm at any time (U.S. EPA 1980b). Copper has been used as algicides and herbicides, because it inhibits photo-synthesis at a concentration as low as 0.001 ppm, and as an antifouling paint on shipping vessels. The elevated levels of copper in Jackson

Ditch sediment may be the result of higher copper in the Gulf Intracoastal Waterway that settles into Jackson Ditch.

Nickel residues in Jackson Ditch sediment were elevated compared to the other locations on the refuge (Table 4). The criterion to protect saltwater aquatic life is 0.001 ppm as a 24-hour average and should not exceed 0.140 ppm at any time (U.S. EPA 1980c). Nickel strongly absorbs to iron and manganese oxides and its toxicity is reduced by calcium and magnesium. Under natural conditions, the nickel present in Jackson Ditch sediment will not be available to aquatic organisms and is not at levels to be of concern at the present time.

Heavy metal residues from the various animals collected on the refuge indicate that only copper and strontium may be accumulating in aquatic species (Table 5). Copper and strontium were the highest in Jackson Ditch sediment and were also high in blue crabs collected from the same area. The mixed aquatic invertebrates collected from the drain ditch of a rice field also had elevated levels of copper and strontium (Table 5). Copper was within the bioconcentration range for these organisms, in areas with elevated sediment levels (U.S. EPA 1980b), suggesting that an equilibrium may have been reached and no further increase in body tissue is expected.

ORGANOCHLORINE PESTICIDES

Eight sediment samples collected from the Anahuac National Wildlife Refuge were analyzed for organochlorines and polychlorinated biphenyls listed in Table 2. None of these contaminants were detected in the sediment samples at the 0.01 ppm detection level (Table 6). These persistent chlorinated compounds are apparently not present, or being deposited, in the sediments of the bayous and irrigation canals on the refuge. Dieldrin and DDE residues were the most frequently detected organochlorines in the biota samples (Table 6), but at levels

See Table/Figure

Table 5. Mean heavy metal residues from various biota collected on the Anahuac National Wildlife Refuge.

(SEE ORIGINAL)

See Table/Figure

Table 6. Organochlorine and polychlorinated biphenyl (PCB) residues detected in samples collected on the Anahuac National Wildlife Refuge in parts per million wet weight.

(SEE ORIGINAL)

below any environmental concerns. The biota samples from both the irrigation canal and the rice farm drainage ditch indicate that the organochlorine residues may be remnants from past agricultural practices. Toxaphene was also recorded at low levels (Table 6) from these two sampling locations. The white crappie sample, which had the highest residue level of 0.96 ppm toxaphene, was below the hazardous range for tertiary consumers (1.7 to 5.5 ppm) reported by Eisler and Jacknow (1985). Toxaphene should not be considered as a contaminant threat to fish and wildlife resources on the Anahuac National Wildlife Refuge, or create a need for a fish advisory.

ORGANOPHOSPHATE PESTICIDES

Organophosphate pesticides are used during the rice growing season both on the refuge and on surrounding farmland. These pesticides have a short half-life after application. Twenty-three organophosphate pesticides were analyzed for in sediment and tissue samples from the irrigation ditch and the drainage canals. No organophosphate was detected in any sample. Use of organophosphate pesticides does occur on the refuge and the fact that they have a short resident time in the environment could be a reason for very little aquatic life (insects, shrimp, tadpoles) present in the rice field drainage ditches for short (i.e., 7 days) periods of time. The subtle ef fects of pesticide use on aquatic life food chain organisms should be investigated in future studies or monitoring efforts.

PETROLEUM HYDROCARBONS

Sediment samples taken from the irrigation canal at the lift pump were contaminated with both aliphatic and polycyclic aromatic hydrocarbons (Table 7). The water pump at this location is usually serviced with diesel fuel and the crank case oil is changed on a regular basis. Spilled oil and grease usually coats the pump and the ground. Waste oil cans and used oil filters are commonly found on the ground around the pump. This heavy contamination by oil at the pump station is the result of sloppy maintenance and servicing of the water pump. The very low presence of petroleum hydrocarbons in the rice field drainage ditch (Table 7) indicates these contaminants are not being carried to a wide area of the refuge.

Jackson Ditch and Oyster Bayou sediment had minor contamination with petroleum hydrocarbons. Both of these sampling areas potentially receive runoff water from petroleum production

facilities. The presence of aliphatic hydrocarbons at a total residue level of 0.340 ppm in the sediment from Jackson Ditch suggests recent contamination. Aliphatic hydrocarbons are toxic to aquatic life at low levels, but they also degrade or volatize

faster than aromatic compounds. This data suggests recent, and perhaps, low level continuous contamination of Jackson Ditch by petroleum hydrocarbons.

Crabs collected from Jackson Ditch had low levels of polycyclic aromatic hydrocarbons in their tissue (Table 8). Other biota collected from the refuge did not have detectable levels of the 14 aromatic hydrocarbons considered in this investigation, except for the three data points in Table 8. The two fish species were collected from the irrigation canal near the lift pump station.

The majority of polycyclic aromatic hydrocarbons entering aquatic environments remain close to the deposition Bite where they accumulate in sediments until benthic organisms either biotransform or biodegrade the compounds (Eisler 1987). Fish and many crustaceans possess the enzymes necessary to activate aromatic compounds during metabolism but most molluscs cannot, which results in bioaccumulation in molluscs (Eisler 1987). Sediments considered as heavily contaminated by polycyclic aromatic hydrocarbons generally contain more than 10 ppm total residues. The 3.6 ppm detected in sediments near the pump station is a moderate contamination, but not at a level that can cause biological effects in aquatic species such as increased liver size in fish, photosynthetic inhibition of macrophytes, and abnormal blood chemistry in molluscs (Eisler 1987).

See Table/Figure

Table 7. Mean petroleum hydrocarbons in two sediment samples collected from each station on the Anahuac National Wildlife Refuge.

(SEE ORIGINAL)

See Table/Figure

Table 8. Mean polycyclic aromatic hydrocarbons in selected biota from Anahuac National Wildlife Refuge. Number of samples in parenthesis.

(SEE ORIGINAL)

CONCLUSIONS

Our preliminary assessment is that the Anahuac National Wildlife Refuge is not contaminated by organochlorine pesticides or heavy

metals at levels that offer any threat to fish or wildlife species. Petroleum hydrocarbons are a continual contaminant problem in small localized areas. Organophosphate pesticides are used on the refuge, and may be responsible for short term loss of aquatic organisms (insect larvae, grass shrimp, tadpoles) in rice fields and drainage ditches. This should be the focus of any future study. Organophosphate residues are difficult to detect in sediments or aquatic tissue.

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APPENDIX I

METAL ANALYSIS

Samples were lyophilized prior to sample digestion. If necessary, the dried sample was then passed through a 2 mm plastic sieve and a split was then ground using a mortar and

pestle. Percent moisture was determined using Standards Methods for the Examination of Water and Wastes, 14th ed. (Section 208A).

Digestions for ICP analysis were performed in accordance with "Procedures for Handling and Chemical Analvsis of Sediment and Water SamDlesll, US EPA/COE, Technical Report EPA/CE-81-1, May 1981. One gram aliquots of the dried samples were digested in a vigorous nitric acid-hydrogen peroxide procedure with a final aqueous matrix dilution of 100 mm after filtration. The sample results are reported in mg/kg dry weight. No extraordinary reactions or color changes were noted for the ICP digestion.

One sample was spiked and duplicated. Summaries of the ICP QC pages follow:

1. Digestion Blanks - Two blanks were digested with the samples. Normal contamination levels for several analytes were found in the blanks.

2. Initial Calibration Checks - The ICP spectrometer was calibrated properly as indicated by the percent recoveries of the elements analyzed (within ten percent windows) in the initial check solutions.

3. Initial Interf erence Check - Background correction factors for selected analytes were properly determined as indicated by percent recoveries for the interference check solutions (within twenty percent windows).

4. Duplicate Analysis - The duplicate precision, as indicated by the Relative Percent Differences (RPD), was acceptable (inside the 20% windows) for all elements with the exception of Al and Pb. Al is only slightly high (21%). The high Pb RPD, at 30%, is probably due to the variability normally found when concentrations are near the IDL.

5. Spike Ana1ysis - Spike recoveries in the sample were within 75 to 125% for most elements. Sb, B, Ag, and Sn were all low. Low recoveries are typically seen for these elements. As a result, the sample results are probably biased low.

6. Ref erence Materials - A solid EPA laboratory control sample (0287) was used as a reference material. Recoveries for certified analyte values which could be quantitated at a level above the reporting limit were all within +/- 25% with the exceptions of Ag. Ag recoveries are typically low with this type of digestion.

ORGANOCHLORINE AND AROCHLOR ANALYSIS

Twenty-four sediment samples were analyzed by Patuxent methods.

A subsample of each well-mixed sediment (5.0 g to 7.3 g), and sodium sulfate (heat treated at 550°C) were blended in a one-half-pint food blender. This mixture was added to a fiber extraction thimble (pre-extracted with petroleum ether). Internal standard solution from a syringe was placed on the sample in the thimble. The sample was extracted with petroleum ether (B&J distilled in glass) for at least 20 hours. The extract was concentrated to 10 mL with a Kuderna-Danish on a steam bath. During the concentration stages, the extract was never allowed to go to dryness.

The 9 mL of extract was exchanged into methylene chloride (Omnisolve distilled in glass) and brought to a 10 mL volume. A volume of extract equivalent to approximately 1 g of sample was loaded into a loop on the GPC unit (ABC model No. 1002A) and injected. The GPC unit transfers the eluted fraction containing the chlorinated organics to an autoconcentrator that concentrates during elution and exchanges the solvent to hexane for a final volume of 10 mL.

The sample was concentrated to 1 mL by nitrogen blowdown and subjected to alumina micro column cleanup. The alumina (Biorad neutral alumina AG7, 100 to 200 mesh) was ignited and then deactivated with distilled water (7% by weight). The analytes were eluted with 10 mL of 4:1 hexane/methylene chloride. The eluent was concentrated to 1 mL for GC capillary analysis.

Percent moisture was determined by placing 2 g of the homogenate into a tared aluminum pan and placed in a drying oven (105°C) for at least 48 hours. The weight was recorded after cooling in a desiccator overnight.

For organochlorine analysis, six chlorinated biphenyl congeners were added before extraction of the sample and served the following purposes:

1. Monitoring sample extract losses due to extraction efficiency, GPC cleanup, or extract transf er.

2. Estimating detection limits.

3. Increasing accuracy of predicted retention times (±0.005 min) for the analytes.

4. Providing backup internal standards in the event of sample matrix interference with the normal quantif ication internal standard.

Before organochlorine GC analysis, two additional internal standards were added to the sample. These were used for monitoring the instrument's health; e.g., to indicate if there were any problems with the injection of each sample.

A Hewlett-Packard 5880A GC equipped with dual capillary column/dual ECD detectors was used for the organochlorine and arochlor analysis. The analysis was a single splitless (Grob) injection onto two 30-meter columns (DB-1 and DB-1701) of different polarities. The dual column analysis, besides providing confirmation of the pesticides, checks for coelution of unknowns with each individual pesticide. Because of the high resolving power of the capillary columns, coelution by an unknown on both columns is improbable. Except as explained below, the amount and variance shown on the sample report pages was calculated from the values given by the two GC columns for each compound detected. If the variance was greater than 15% of the mean, it was assumed that coelution was occurring on the column showing the higher amount and only the lower amount was reported. In that case, a variance indicator NA (Not Applicable) was printed in the "Variance" list. Also, if near coelution occurs, where a positive identification on one of the GC columns was not possible, then only the amount given by the GC column that allows positive identification was reported. In this case, the variance indicator NA also was printed. The indicator NA also was used in the "Variance" list in cases where nothing was found above the detection limits on either column where the indicator ND was printed in the "Amount" list.

The temperature program was 500C for two minutes to 280°C at 3°C/minute and a post-run temperature of 290°C for five minutes. Thinear flow rate was at 30 cm helium/second.

Quantitation was done on the Hewlett-Packard 5880A GC. Due to the narrowness of the capillary peaks, all data were based on peak height, resulting in less biasing due to tailing, near coelution and baseline drift ("Assessment of the Results from Data Processing Svstems using a Digital Chromatogram Simulator", R.J. Hunt, Journal of High Resolution Chromatography Communica-tions, Vol. 8, July 1985, pp. 347-355). All data were collected directly from the GC into databases in an Amiga computer. The databases, besides providing report generation, allow the monitoring of the standard curves and internal standards over time. The data on the Amiga also was used for pattern recognition in arochlor analysis and to develop the organo chlorine pesticide "unknowns" report. Appendices A and B contain the results of the organochlorine-arochlor and "unknowns" analyses, respectively.

The batch size for soxhlet extraction was 12 (11 samples and 1 blank). Two batches went onto the GPC at a time. No analytes

were detected in the blank at concentrations >0.5 ppb.

No GC/MS confirmation was done since no analytes were detected.

ALKANE AND AROMATIC ANALYSIS

Sample preparation for the alkanes and aromatics was as follows. Five micrograms deuterium labeled surrogate spikes were added to 5-15 g of the sample homogenate. There were labeled analogs for each of the polyaromatic hydrocarbons to be analyzed except benzo(e)pyrene and perylene. Aqueous potassium hydroxide (4 N) was added to each of the mixtures and the sample saponif ied in a steam bath for two hours. The centrifuge tubes were vortex mixed every 40 minutes. The hydrolysates were acetif ied with hydrochloric acid, the mixture transferred to a separatory funnel and extracted three times with 25 mL methylene chloride each time. The aqueous layer was discarded. Soil and sediment samples were not hydrolyzed. The samples were mixed with sodium sulfate and soxhlet-extracted overnight with methylene chloride. The combined organic extract filtered through muf f led NA2SO4 and rotary-evaporated to several millimeters. One hundred mL petroleum ether and 0.7 mL iso-octane was added prior to initial evaporation and the extract again reduced to several millimeters.

The alkanes and aromatics were fractionated on a column of 20 g 2.0% water-deactivated silica gel. Alkanes were eluted with 100 mL 40% methylene chloride in petroleum ether and an additional 60 mL methylene chloride. Each fraction was concentrated by rotary evaporation followed by nitrogen evaporation. The alkane fraction was evaporated to 1 mL, internal standards added and the extract transferred to a vial in preparation for GC analysis.

The aromatic fraction was concentrated to 10 mL and cleaned by gel permeation chromatography on Bio-Beads SX-3. The collected gel permeation fraction was first rotary-evaporated, then nitrogen-evaporated to 1 mL and finally shaken with aqueous sodium hydroxide. This step removed residual fatty acids. An injection internal standard was added to each extract and it was transferred to a vial in preparation for GC analysis.

Three compounds, n-undecane, n-docosane, and n-triacontane were added to each of the final alkane extracts before GC analysis to serve as quantitation internal standards.

Gas chromatography was done using a 30 M DB-5 capillary column with splitless injection on a Hewlett-Packard 5880A GC with flame ionization. The temperature program was 60°C for three minutes to 310°C at 60/minute for alkanes and a post run temperature of

320°C for two minutes. Linear flow rate was 30 cm helium/second.

Internal standards for the polyaromatic hydrocarbons were the deuterium labeled compounds added at the saponification stage. The deuterium labeled fluorene has been found to deuterium/hydrogen exchange during base hydrolysis. Thus, D[sub]10 phenanthrene was used as the internal standard for fluorene.

Use of these internal standards automatically compensates for any losses during sample preparation. An injection internal standard was added to each extract before analysis on the GC/MS and was used to determine if recovery of labeled compounds were within the normal expected range.

Gas chromatography was done using a 30 M DB-5 capillary column with splitless injection on a Hewlett-Packard 5890 GC in conjunction with a Finnigan-MAT INCOS 50 mass spectrometer. The temperature program was 50°C for two minutes to 320°C at 8°/minute. The mass spectrometer scanned from 35 to 450 m/z in 0.56 seconds at 70 eV.

The target polyaromatic hydrocarbons were purchased from Supelco (Supelpreme) and mixtures of isotope labeled compounds were purchased from MSD Isotopes. Responses of the labeled compounds to 2,2'-dif luorobiphenyl internal standard and of the target to the labeled compounds was used to create a polyaromatic hydrocarbon library response list. The response curves for the target polyaromatic hydrocarbons were generated from 1 to 50 ng on column and were linear in this range.

The mass spectrometer was calibrated and an on-going calibration verification standard at either 1 or 2 ng on column injected daily. Compounds were searched for and quantif ied with "TCA", a program available from Finnigan-MAT for the analysis of target compounds. Mass spectra were examined manually to verify identification.