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DETERMINATION OF CADMIUM, CHROMIUM, NICKEL, AND LEAD IN URINE USING GRAPHITE FURNACE ATOMIC ABSORPTION SPECTROPHOTOMETRY

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K-25 Technical Services Division

Sponsored by R. W. Morrow

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ABSTRACT

Procedures using graphite furnace atomic absorption spectrophotometry for the determination of cadmium, chromium, nickel, and lead are described in this report. The lowest concentrations reported using the procedure are 5 μg/l for cadmium, chromium, and nickel and 10 μg/l for lead.

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DETERMINATION OF CADMIUM, CHROMIUM, NICKEL, AND LEAD IN URINE USING GRAPHITE FURNACE ATOMIC ABSORPTION SPECTROMETRY

SUMMARY

Procedures have been developed to determine cadmium, chromium, nickel, and lead in urine using graphite furnace atomic absorption spectrophotometry. Cadmium, nickel, and lead are determined after being extracted from urine as the ammonium pyrrolidine dithiocarbamate complex using methyl isobutyl ketone (MIBK) as the solvent. The metal concentration of the extracts is determined by the method of standard additions using an atomic absorption spectrophotometer equipped with a deuterium lamp for background correction. Chromium is determined directly in the urine sample by the method of standard additions and requires an atomic absorption spectrophotometer which is equipped with a tungsten-halogen background corrector. The lowest concentrations reported are: 5 μg/l for cadmium, chromium, and nickel, and 10 μg/l for lead. Precision expressed as standard deviation is as follows:

0.5 μg/l for cadmium determined at 5 μg/l,

0.8 μg/l for chromium determined at 5 μg/l,

1.2 μg/l for nickel determined at 10 μg/l, and

1.7 μg/l for lead determined at 10 μg/l.

The detailed procedure for determining these elements in urine is presented in the Appendix.

INTRODUCTION

The Industrial Hygiene Department at the Oak Ridge Gaseous Diffusion Plant (ORGDP) routinely submits urine samples for the determination of chromium, nickel, and lead. These samples are used to monitor employees who might have received exposure to these metals in their work environment.

Flame atomic absorption has frequently been used to determine trace metals in urine samples, but lacks the sensitivity necessary to detect low exposure levels. However, more recently Graphite Furnace-Atomic Absorption (GFAA) spectrophotometry has become a routine analytical tool in many laboratories. This method has μg/l sensitivity for many toxic metals of industrial hygiene interest. There have been published reports concerning the use of this technique for determining various elements in urine.(1-6) Advantages of the graphite furnace technique include the following: ease of sample preparation, increased sensitivity, and smaller sample required.

The major difficulty with atomic absorption measurements for the determination of metals in urine has been the necessity for accurate and reproducible background correction during atomization.(5) The installation in the ORGDP laboratory of an automated Perkin-Elmer Model 5000/HGA-500 atomic absorption-graphite furnace instrument with its improved background correction system has permitted corrections to be made for all elements. A major improvement in the Model 5000 is the tungsten iodide source for background correction at wavelengths greater than 300 nm, whereas, older systems permitted corrections to be made only for wavelengths in the ultraviolet region.(3) The Perkin-Elmer Model 5000/HGA-500 system also permits the automated sequential analysis of up to six elements in up to thirteen samples.

This report concerns the study undertaken in the ORGDP laboratory to optimize conditions and to evaluate the graphite furnace technique for determining cadmium, chromium, nickel, and lead in urine samples. The technique was found to be ideally suited for automatically determining these elements using the Perkin-Elmer Model 5000 atomic absorption/500-graphite furnace system. A solvent extraction technique is used in the determination of cadmium, nickel, and lead, whereas, chromium is determined directly using the acidified sample. The methods-of-additions technique is used for all elements to minimize matrix effects.

ANALYSIS OF URINE FOR CADMIUM, CHROMIUM, NICKEL, AND LEAD

EXPERIMENTAL

Instrumentation and Apparatus

All measurements were made with a Model 5000 atomic absorption spectrophotometer (shown in Figure 1) which utilizes these two continuum sources for background correction; a deuterium arc lamp for the ultraviolet region and a tungsten iodide source for the visible region of the spectrum.(7) The instrument was equipped with a Model HGA-500 Graphite Furnace which utilizes a temperature ramp accessory,(8) a Model 056 recorder, and a Model AS-40 Auto Sampling System for the graphite furnace. (All instruments were from Perkin-Elmer Corporation, Norwalk, Connecticut 06856.)

The Model 5000 is provided with microcomputer electronics which monitor and control every instrumental operation. The instrument coupled with the automatic card reader and the auto sampler accessories permit automated sequential multielement analyses to be performed for six elements in up to 35 samples. The magnetic card reader attachment permits the storage of analytical conditions for up to six elements on a magnetic card. The six analytical programs can be recalled, when needed, from the card and transferred to the instrument's internal program memory.

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Figure 1
PERKIN-ELMER MODEL 5000 ATOMIC ABSORPTION SPECTROPHOTOMETER

The HGA-500 graphite furnace is also microcomputer-controlled which permits the use of up to nine program steps for each determination. The furnace also permits the storage of up to six programs in its internal memory for recall and use at any time, and programs can also be stored with an optional magnetic card reader. The AS-40 auto sampler consists of a microcomputer-controlled programmer and a sampler. Combined with the Model 5000 atomic absorption spectrophotometer and the HGA-500 Graphite Furnace, the AS-40 is able to perform sequential determinations of up to six different elements in up to 35 samples without operator attention, permitting a truly automated operation. The instrument conditions used for this study are shown in Table 1.

Before initial use, glassware is soaked in 10% nitric acid and rinsed three times with deionized distilled water. This glassware is used exclusively for the analysis of trace metals in urine. Blank analyses are made on all glassware, and no significant contamination has been encountered. When operating the instrument manually, Eppendorff microliter pipettes with disposable plastic tips are used for the injection of the sample into the furnace. The tips are rinsed three or four times with the solution to be injected in order to eliminate contamination. Pyrolytically coated graphite tubes are used and to prevent contamination, each new tube is taken through the sample heating sequence on the graphite furnace until all of the element of interest is removed. The pyrolytic tubes have been found to be usable for approximately 100 firings.

Reagents

All reagents used were at least ACS reagent grade. The deionized distilled water that was used was prepared by passing distilled water through a mixed bed of cation and anion exchange resin. The ammonium pyrrolidine dithiocarbamate (APDC) solution, 2% v/v, was prepared by dissolution in deionized distilled water followed by extraction with methyl isobutyl ketone (MIBK) to remove impurities. Commercial, certified standards (Fisher Scientific Company) were used for the cadmium, chromium, nickel, and lead stock solutions. High purity ("Ultrex" from J. T. Baker Chemical Company) nitric and acetic acids were used for the acidification of the samples and aqueous standards.

Sample Preparation Procedure

All urine samples were acidified immediately upon receipt in the laboratory to 1% (v/v) with acetic acid to preserve the sample and the method-of-additions technique was used for all samples.

Preparation Procedure for Cadmium, Lead, and Nickel.

(1) Pipet 5 ml of urine into each of three 15-ml cap screw centrifuge tubes. Label these tubes Nos. 1, 2, and 3, respectively.

(2) To Tube No. 1, add 1 ml deionized distilled water. To Tube No. 2, add 1 ml of 0.025 μg/ml cadmium or 1 ml of 0.05 μg/ml nickel or lead standards. To Tube No. 3, add 1 ml of 0.05 μg/ml cadmium or 1 ml of 0.1 μg/ml nickel or lead standards. Mix each sample with a Vortex mixer.

Table 1

INSTRUMENT PARAMETERS USED FOR DETERMINING
CADMIUM, CHROMIUM, NICKEL, AND LEAD IN URINE

Element	Cadmium	Chromium	Nickel	Lead
Wavelength (nm)	228.8	357.9	232.0	283.3
Slit Setting (nm)	0.7	0.7	0.2	0.7
Mode	Absorbance	Absorbance	Absorbance	Absorbance
Signal	Peak Height	Peak Height	Peak Height	Peak Height
Background Correction	Deuterium Arc	Tungsten-Halogen	Deuterium Arc	Deuterium Arc
Purge Gas	Argon	Argon	Argon	Argon
Gas Interrupt	No	Yes	No	Yes
Dry (temp/total time)	130°/30 sec	105°/100 sec	130°/20 sec	130°/30 sec
Char (temp/total time)	350°/30 sec	1,300°/240 sec	900°/40 sec	600°/50 sec
Char Ramp Time	10 sec	120 sec	10 sec	20 sec
Atomize (temp/total time)	2,000°/8 sec	2,700°/10 sec	2,700°/10 sec	2,300°/10 sec
Calibration	Standard Additions	Standard Additions	Standard Additions	Standard Additions
Sample Size	20 μl	20 μl	20 μl	20 μl

(3) Adjust the pH of each solution to 2.8 ± 0.2 with 1.0 N HCL. Mix with the Vortex mixer.

(4) Add 0.2 ml of the APDC solution to each tube and mix thoroughly.

(5) Pipet 5.0 ml of MIBK into each tube. Cap with Teflon-lined caps and shake for 2 min.

(6) Centrifuge (2,800 rpm) for 10 min.

Preparation Procedure for Chromium.

(1) Pipet 5 ml of urine into each of three 15-ml centrifuge tubes. Label these tubes Nos. 1, 2, and 3 respectively.

(2) To Tube No. 1, add 1 ml deionized distilled water. To Tube No. 2, add 1 ml of 0.025 μg/ml chromium standard. To Tube No. 3, add 1 ml of 0.05 μg/ml chromium standard. Mix each with a Vortex mixer.

Analysis Procedure

Manual Operation. Adjust the instrument to the appropriate conditions for the element being determined as shown in Table 1. (These parameters were optimized for each element using the technique described in the Perkin-Elmer Analytical Methods Manual.(2,7,8) Using an Eppendorff microliter pipette, inject 20 μl of the appropriately prepared sample or sample plus standard into the sample injection port of the graphite furnace. Initiate the programmer on the graphite furnace to begin the dry, char, and atomize cycle. Record the absorbance reading and from the absorbance data of the sample and the sample plus added standards, plot a "standards additions" curve. Read the sample concentration from the curve. Typical "methods-of-additions" plots obtained on samples are shown in Figures 2 and 3.

Automatic Operation. Turn on the atomic absorption spectrophotometer and, using the magnetic storage cards, enter into the spectrophotometer programmer the operating parameters for the elements to be determined. Turn on the graphite furnace programmer and, using the magnetic cards, enter into the furnace programmer the operating parameters for the elements to be determined. Align the sampling capillary tip of the AS-40 auto sampler with the sample introduction hole in the graphite tube. Place the standard and sample solution in the sample tray, and place the tray on the trough, and cover. Enter the concentration of the standard solution used to spike the samples into the spectrophotometer, and enter the required volume and program for the element into the AS-40 programmer. If additional elements are to be determined in the sample, enter the proper sequence keys and enter the required volume and program parameters for each element. Initiate the analysis run sequence; each sample is then analyzed in turn, and the results printed out on an associated printer.

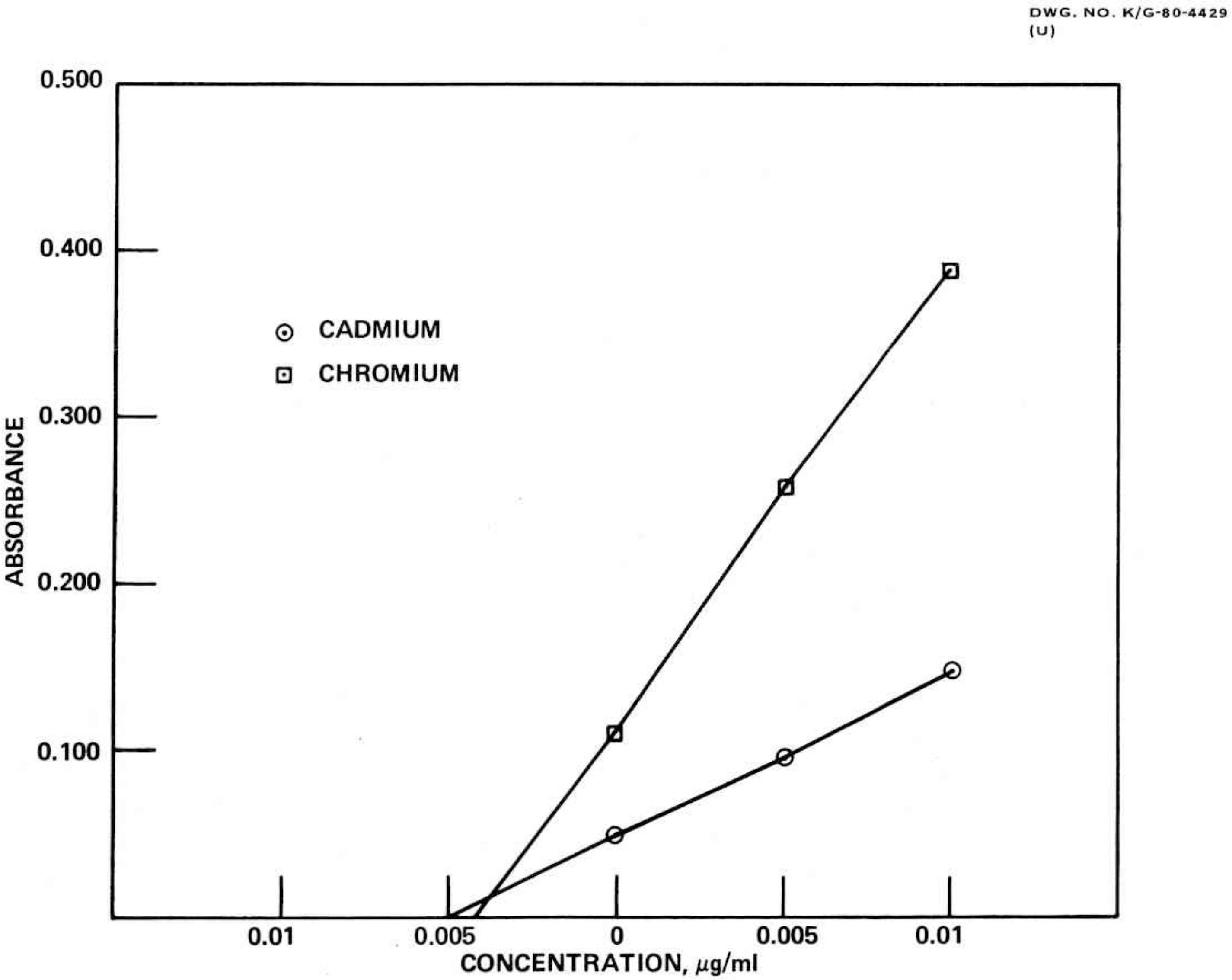

Figure 2
METHOD OF ADDITION PLOTS ON SAMPLES

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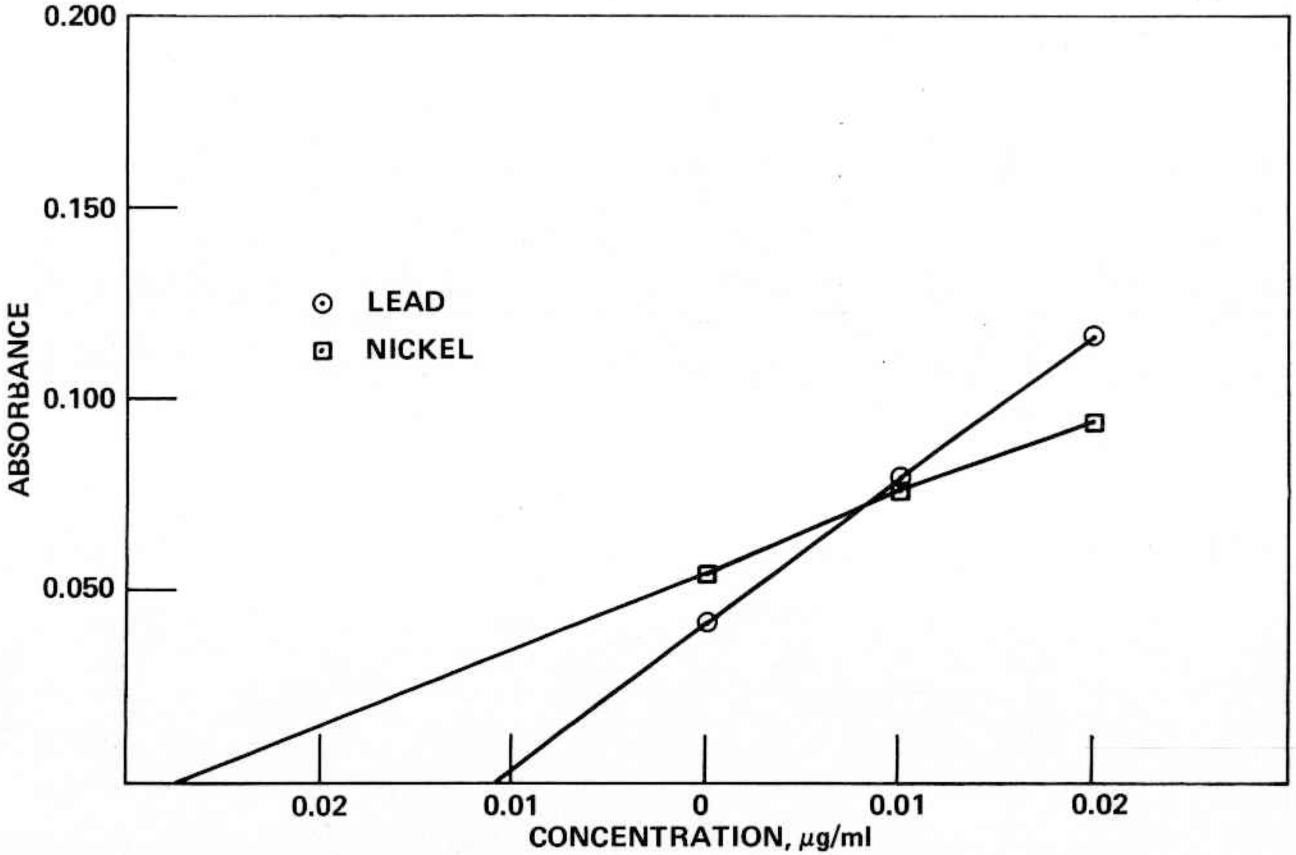

Figure 3

METHOD OF ADDITION PLOTS ON SAMPLES

RESULTS AND DISCUSSION

Cadmium, Nickel, and Lead

An ammonium pyrrolidine dithiocarbamate (APDC)-methyl isobutyl ketone (MIBK) extraction procedure was used for the determination of cadmium, lead, and nickel. It had been reported that analytical techniques which involved extraction steps to separate the element from the urine matrix prior to flameless atomic absorption spectrophotometry were generally superior to techniques which involve the direct analysis of urine.(6) The extraction procedure of Willis(9) and Berman(10) and as used by Morrow and McElhaney(1) was chosen. APDC is a group reagent which will complex all three of the desired elements at a common pH. Figures 4, 5, and 6 show calibration curves prepared from extracting aqueous standards and from extracting spiked urine standards for all three elements. The slope of the curve for both nickel and lead prepared from the spiked urine is somewhat less than that of the curve prepared from the aqueous standards, whereas, the slopes of the curves for cadmium are the same. Therefore, the method of standard additions was chosen as the method of calibration to minimize any possible matrix interferences.

To check the cadmium, nickel, and lead stability in urine samples before extraction, two 100-ml flasks of urine were spiked to contain 0.01 μg/ml cadmium and 0.03 μg/ml of both nickel and lead. Flask 1 contained 1% acetic acid and Flask 2 contained no acid. These flasks were stored at room temperature, and aliquots from each flask were extracted daily. The results of this study are shown in Table 2. The study shows that if lead or cadmium are to be determined, the sample should be acidified immediately upon receipt to prevent loss of these elements. After acidification, the samples are stable for 2 days. The study also shows nickel is stable in the "as received" sample for 3 days and is stable in the acidified sample for at least 5 days.

Table 2

STABILITY OF TRACE METALS IN URINE - NO ACID AND 1% CH_3COOH

Analysis	0.01 μg Cd/ml		0.03 μg Ni/ml		0.03 μg Pb/ml	
Day	No Acid	1% CH_3COOH	No Acid	1% CH_3COOH	No Acid	1% CH_3COOH
1	0.012	0.011	0.038	0.034	0.033	0.031
2	0.004	0.009	0.032	0.034	0.000	0.032
3	0.001	0.007	0.034	0.034	0.001	0.020
4	0.001	0.004	0.024	0.034	0.001	0.014
5	0.001	0.003	0.018	0.032	0.000	0.009

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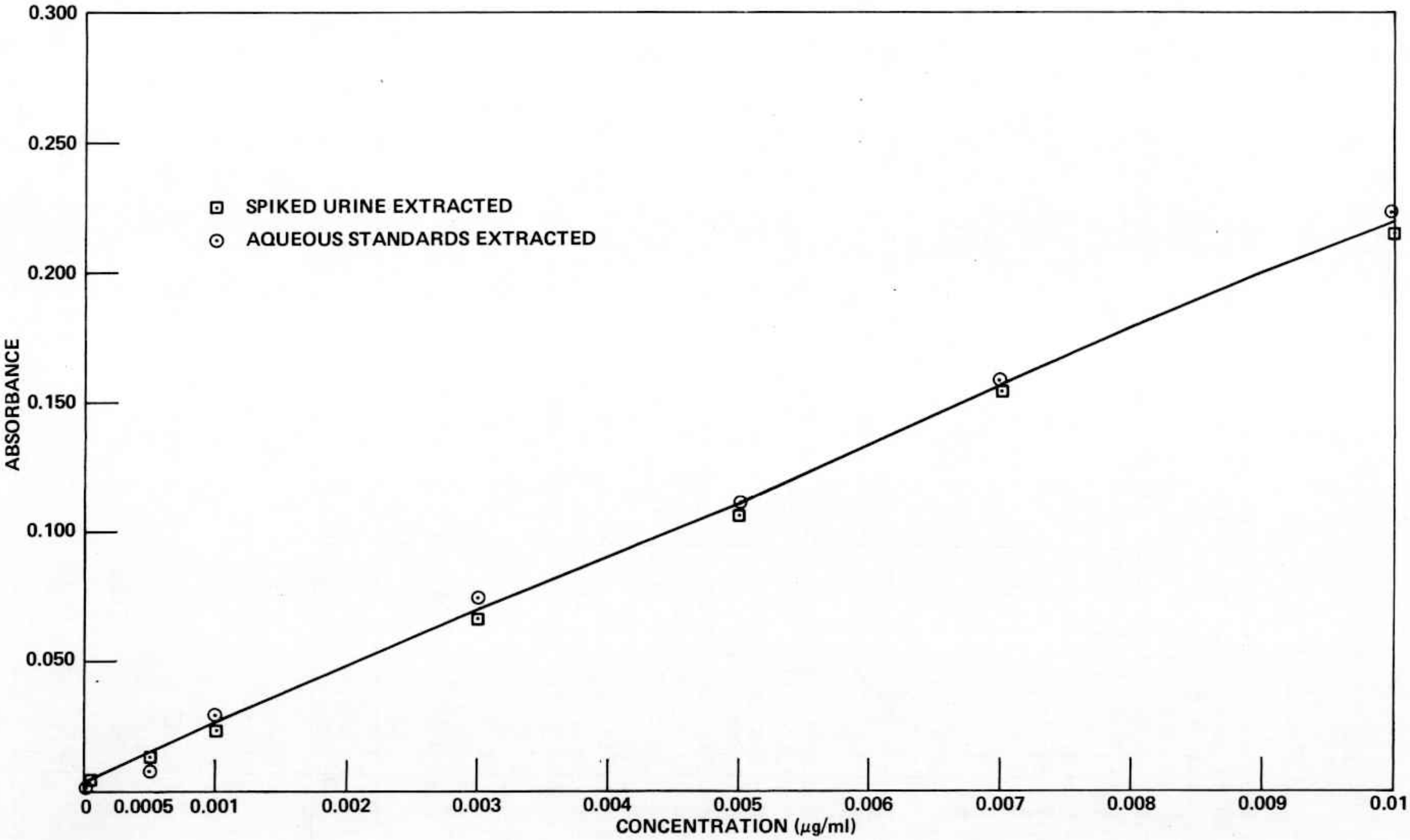

Figure 4
CADMIUM (MiBK EXTRACTED)

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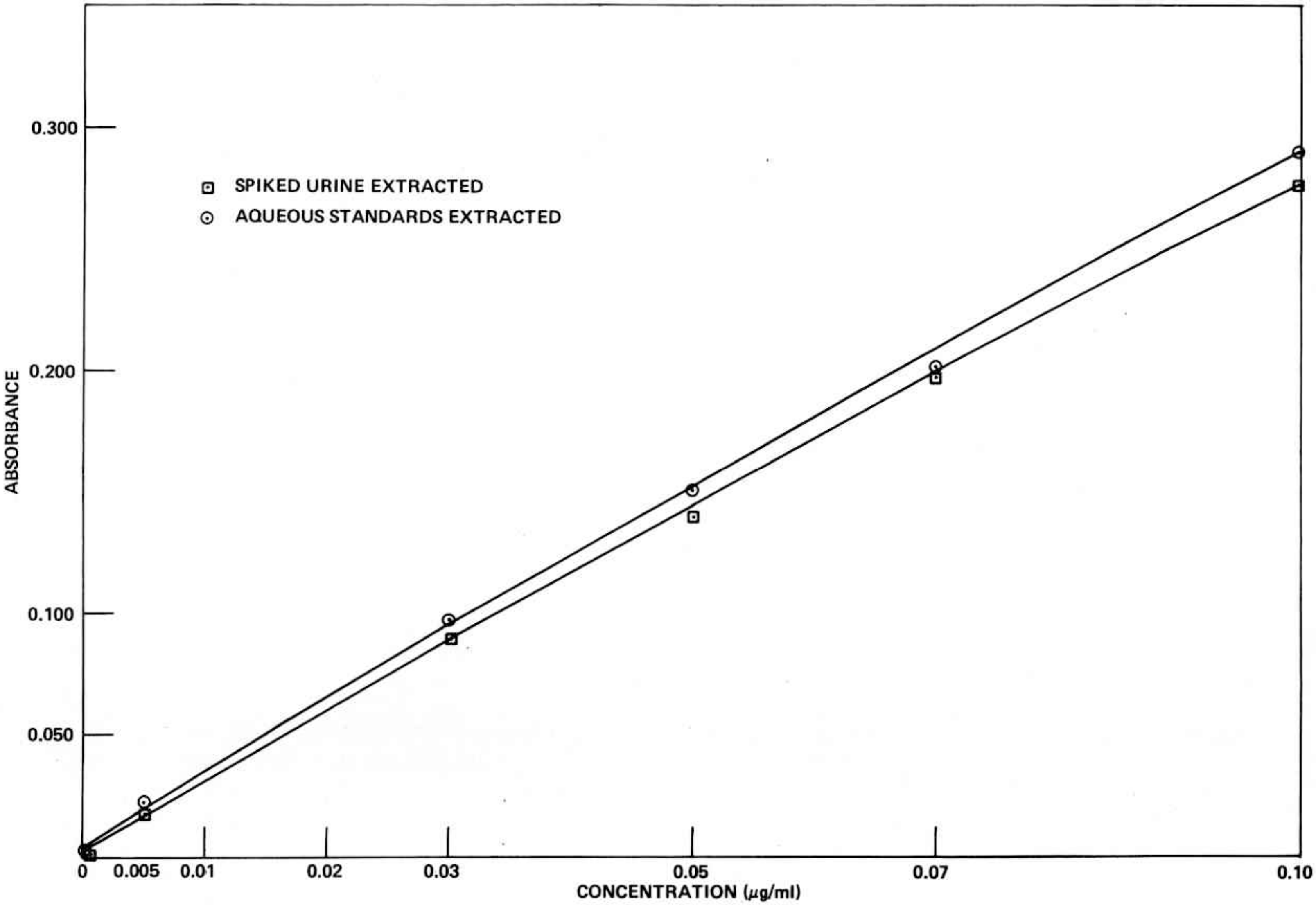

Figure 5
Ni (MiBK EXTRACTED)

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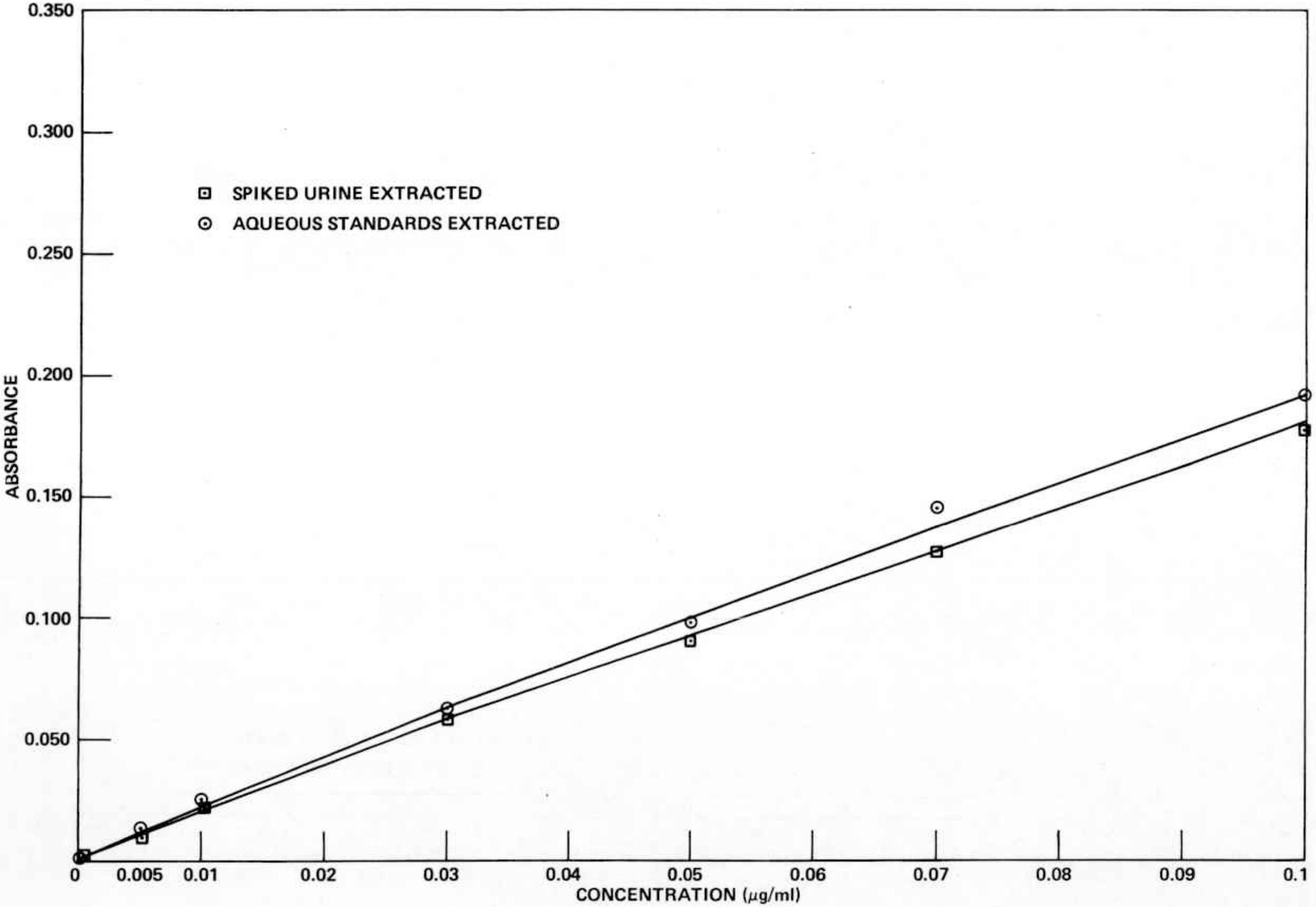

Figure 6
Pb (MiBK EXTRACTED)

To check the cadmium, nickel, and lead stability in the MIBK after extraction, urine samples were spiked to contain 0.01 μg/ml cadmium and 0.03 μg/ml of nickel and lead and taken through the extraction procedure. The extracted samples were centrifuged to separate the organic and aqueous layers, but were not decanted. The extracted samples were kept in a closed container at room temperature and analyzed at appropriate intervals for the desired element. The results are shown in Figures 7, 8, and 9. The results indicate that nickel is stable for approximately 2 days after extraction, however, cadmium should be analyzed within approximately 7 hr of being extracted and lead within 5 hr.

Chromium

One of the major difficulties in measuring chromium in urine by furnace atomic absorption has been the lack of adequate background correction during atomization. The Perkin-Elmer Model 5000 atomic absorption spectrophotometer is equipped with a background correction system which provides simultaneous, continuum source background correction for all elements. This system permits chromium to be determined in urine directly using the method-of-additions technique. Furnace conditions were optimized from conditions described by Perkin-Elmer Corporation in their Analytical Manual using the HGA Graphite Furnace.(2) Similar conditions have been reported in other studies.(3,4,5) The long char time with the long char ramp time used in the procedure is necessary to eliminate matrix interferences.

A urine sample was spiked and analyzed and the resulting analytical curve was compared in Figure 10 with the analytical curve obtained from aqueous standards. The slopes of the resulting curves were different, thus indicating that the method of additions is necessary for correction of matrix effects.

To check the chromium stability in urine samples, three samples were spiked to contain 0.005 μg/ml chromium. One sample was not acidified while one sample was prepared to contain 1% nitric acid and one to contain 1% acetic acid. These samples were analyzed over a period of 16 days; and the results, shown in Table 3, indicate the chromium is stable in the "as received" sample as well as in the acidified sample for this period. The results, obtained on the acidified samples were less erratic than those obtained on the non-acidified sample, therefore, the sample should be acidified upon receipt in the laboratory.

Sensitivities and Detection Limits

The sensitivities and detection limits for all four of the elements determined using the procedure described are listed in Table 4. Sensitivity for the elements were computed from the slope of the plot of percent absorption as a function of concentration. Detection limit is defined as the concentration of the element necessary to yield an absorption signal twice the standard deviation of the background.

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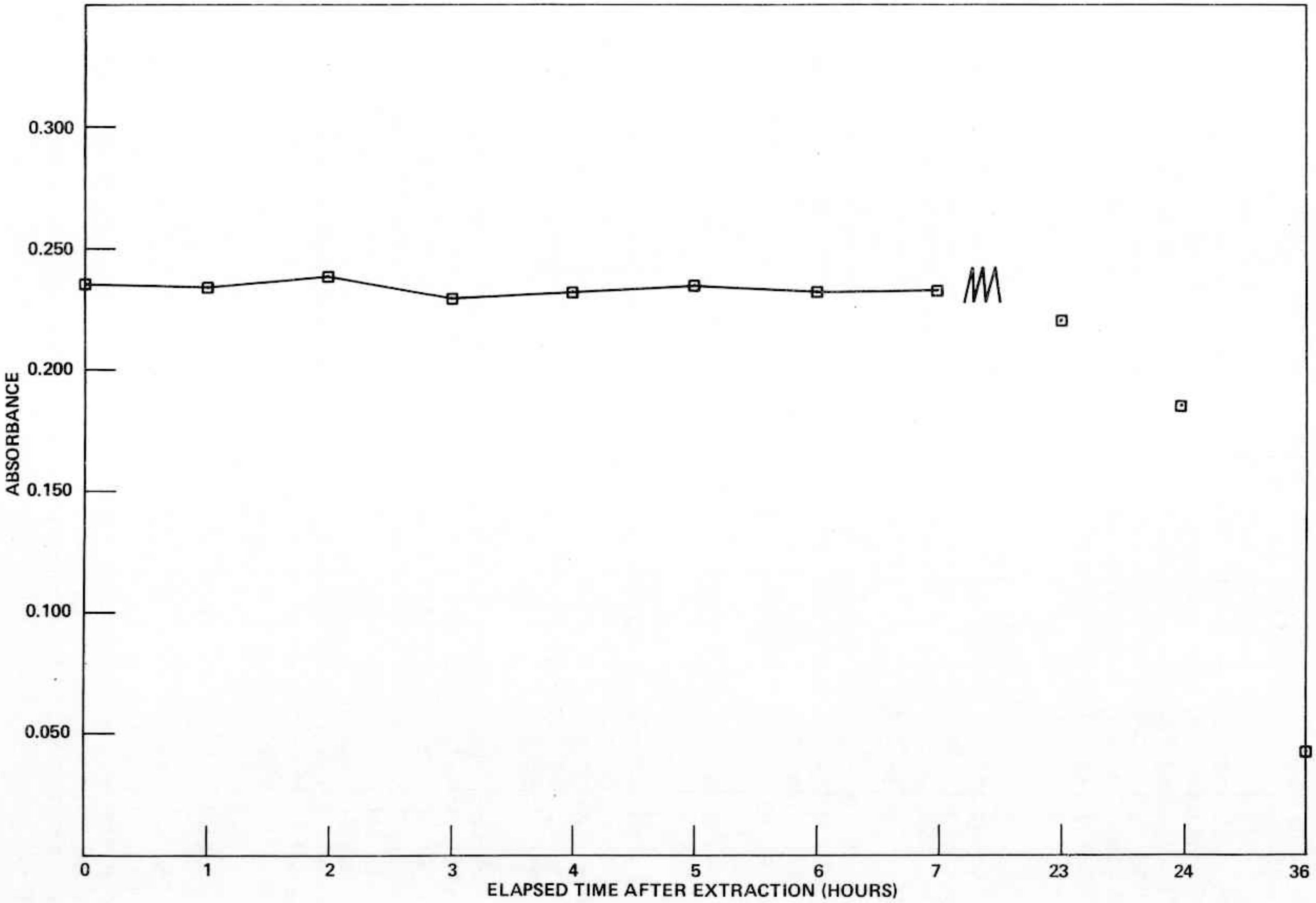

Figure 7

STABILITY OF CADMIUM AFTER EXTRACTION

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(U)

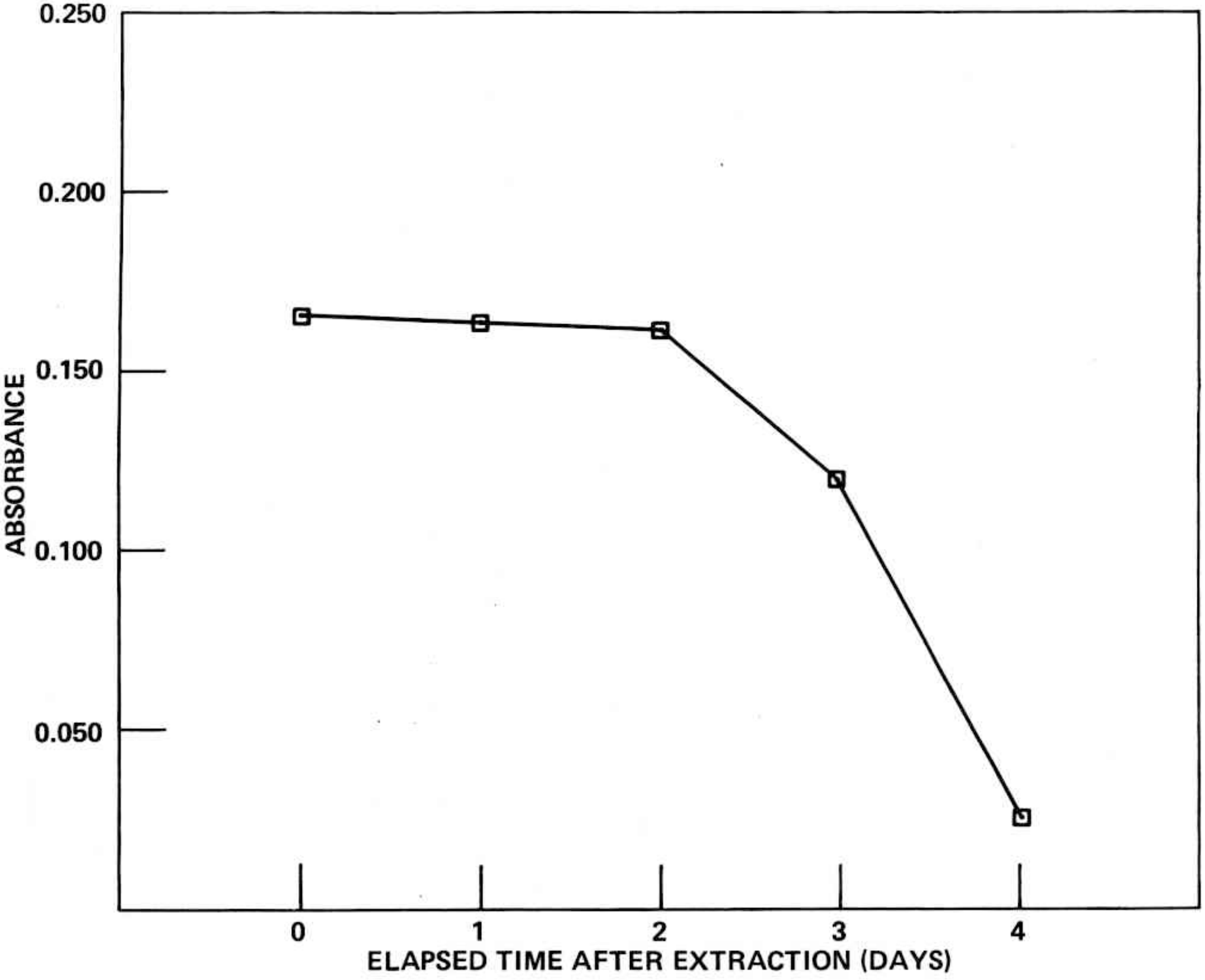

Figure 8
STABILITY OF NICKEL AFTER EXTRACTION

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(U)

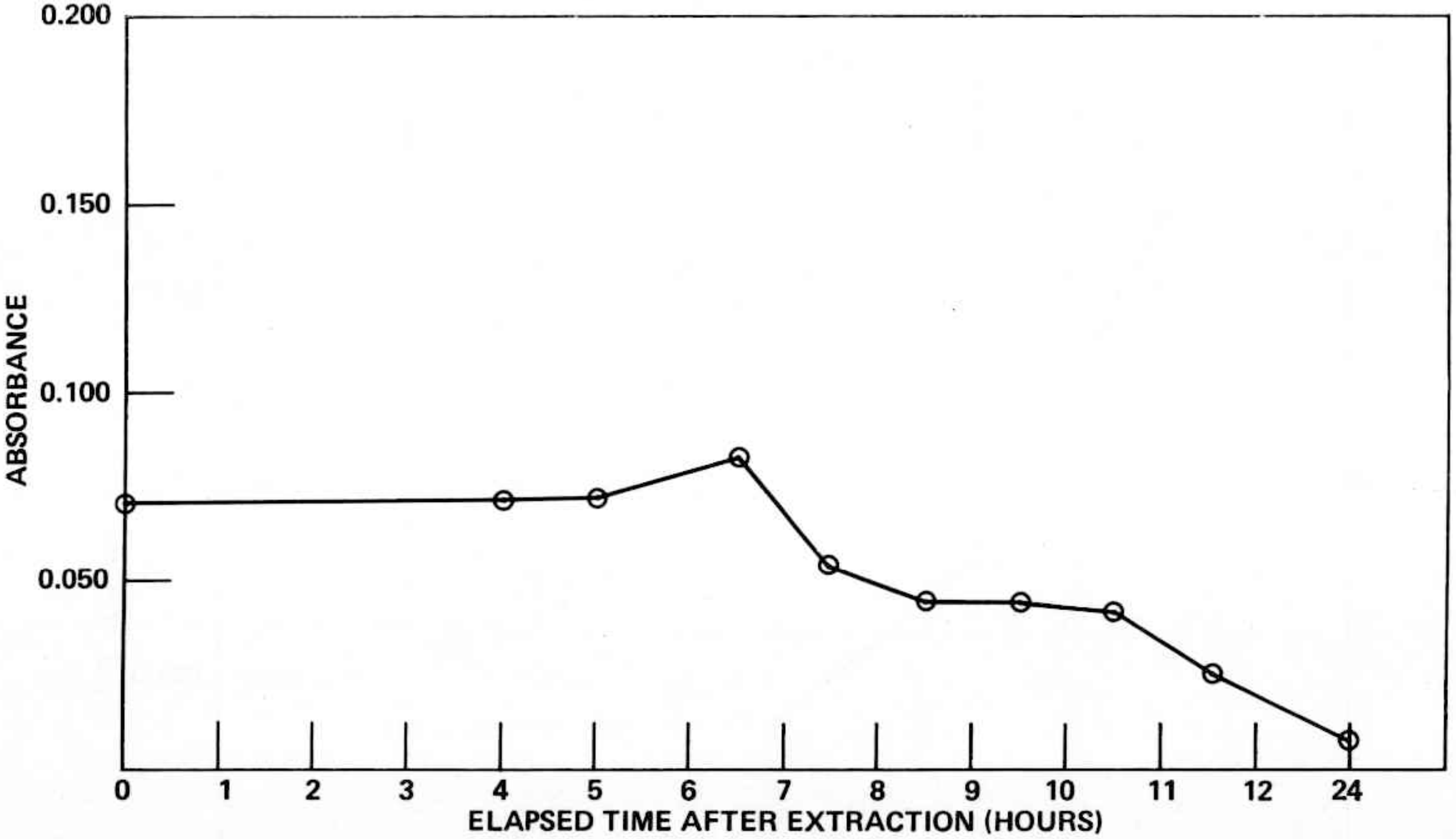

Figure 9

STABILITY OF Pb AFTER EXTRACTION

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(U)

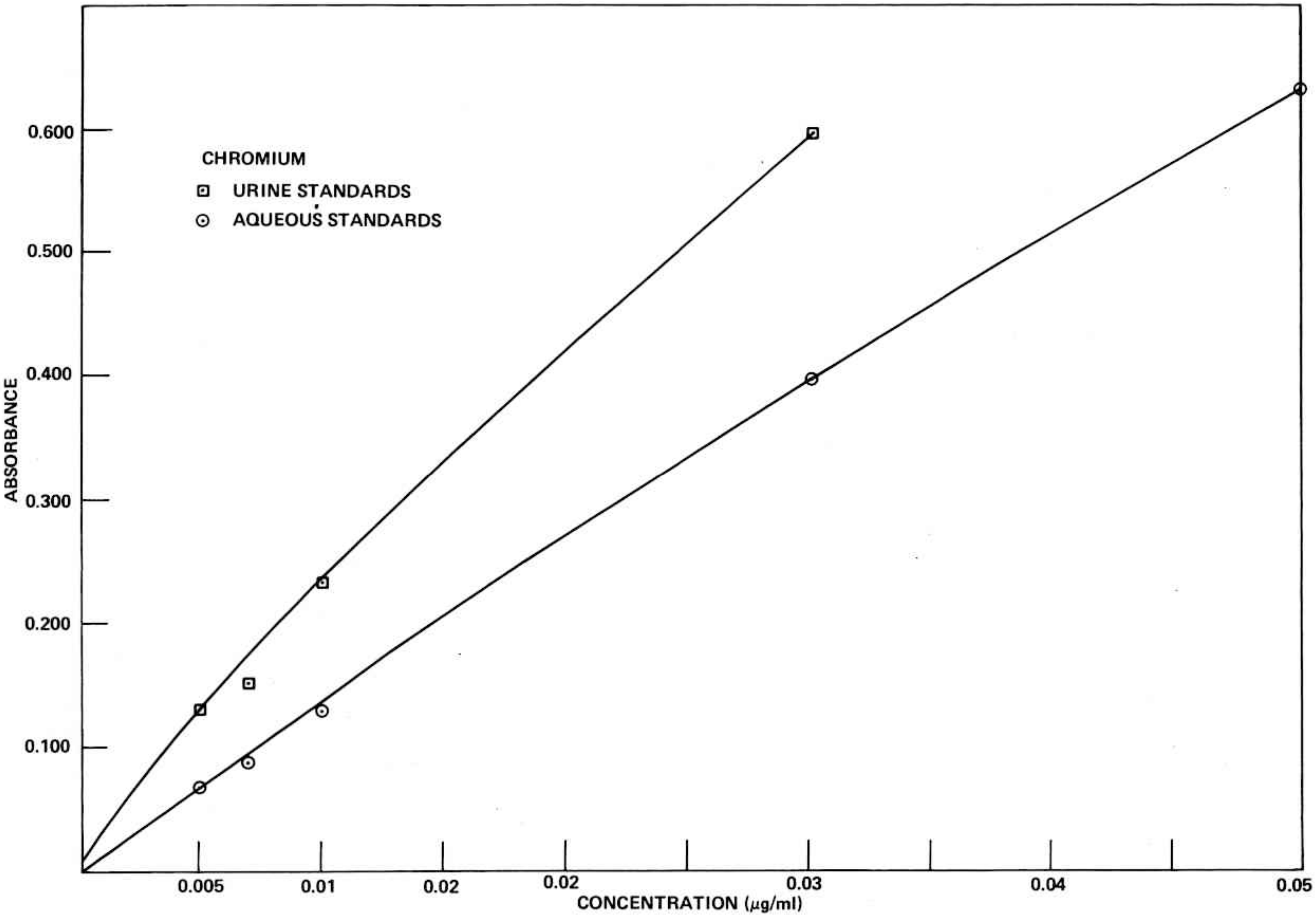

Figure 10
ANALYTICAL CURVES – CHROMIUM

Table 3

STABILITY OF CHROMIUM IN URINE USING DIFFERENT ACIDS

Analysis Day	0.005 μg/ml Cr Added 1% HNO_3 μg/ml	1% CH_3COOH μg/ml	No Acid μg/ml
1	0.005	0.004	0.005
2	0.005	0.004	0.004
3	0.004	0.004	0.006
4	0.005	0.005	0.005
7	0.005	0.005	0.005
9	0.005	0.005	0.006
10	0.005	0.005	0.007
11	0.005	0.005	0.007
15	0.005	0.005	0.006
16	0.005	0.005	0.006

Table 4

SENSITIVITY AND DETECTION LIMIT

Element	Sensitivity (μg/l/1% A)	Detection Limit μg/l
Cadmium	0.18	1.1
Chromium	0.20	1.7
Nickel	1.96	2.5
Lead	4.34	3.1

Precision and Accuracy

The day-to-day precision and accuracy of the technique are reported in Table 5. These data were taken from spiked urine samples and analyzed as samples.

Table 5

PRECISION OF METHOD

	Mean, $\mu g/l$	% R.S.D.
Cadmium	6.5	8
Chromium	5.6	15
Nickel	10.0	12
Lead	8.8	17

REFERENCES

1. McElhaney, R. J. and Morrow, R. W., *Determining Lead in Urine by Flameless Atomic Absorption Spectroscopy,* Union Carbide Corporation, Nuclear Division, Y-12 Plant, Oak Ridge, Tennessee, Y-DK-90 (1972).

2. Perkin-Elmer Corporation, *Analytical Methods Using the HGA Graphite Furnace,* Norwalk, Connecticut (1977).

3. Kayne, F. J., Komar, G., Laboda, H., and Vanderlinde, R. E., "Atomic Absorption Spectrophotometry of Chromium in Serum and Urine with a Modified Perkin-Elmer 603 Atomic Absorption Spectrophotometer," *Clinical Chemistry,* Vol. 24, No. 12, pp 2151-2154 (1978).

4. Routh, M. W., "Analytical Parameters for Determination of Chromium in Urine by Electrothermal Atomic Absorption Spectrometry," *Analytical Chemistry,* Vol. 52, No. 1, pp 182-185 (1979).

5. Guthrie, B. E., Wolf, W. R., and Veillon, C., "Background Correction and Related Problems in the Determination of Chromium in Urine by Graphite Furnace Atomic Absorption Spectrometry," *Analytical Chemistry,* Vol. 50, No. 13, pp 1900-1902 (1978).

6. Adams, D. B., Brown, S. S., Sunderman, F. W., Jr., and Zachariasen, H., "Interlaboratory Comparisons of Nickel Analyses in Urine by Atomic Absorption Spectrometry," *Clinical Chemistry,* Vol. 24, No. 6, pp 862-867 (1978).

7. Perkin-Elmer Corporation, *Instructions - Model 5000 Atomic Absorption Spectrophotometer,* Norwalk, Connecticut (1979).

8. Perkin-Elmer Corporation, *Instructions - Model HGA 500 Graphite Furnace,* Norwalk, Connecticut (1978).

9. Willis, J. B., "Determination of Lead and Other Heavy Metals in Urine by Atomic Absorption Spectroscopy," *Analytical Chemistry,* Vol. 34, pp 614-617 (1962).

10. Berman, E., "The Determination of Lead in Blood and Urine by Atomic Absorption Spectrophotometry," *Atomic Absorption Newsletter,* Vol. 3, pp 111-114 (1964).

APPENDIX

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APPENDIX

Principle of the Method

1. Cadmium, Lead, and Nickel

    a. The elements are extracted from the urine sample as the ammonium pyrrolidine dithiocarbamate (APDC) complex into methyl isobutyl ketone (MIBK).

    b. The metal concentration is determined using the methods of standard additions by atomic absorption spectroscopy using a graphite furnace.

2. Chromium

    a. Chromium is determined directly in the urine sample using the methods of standard additions by atomic absorption spectroscopy using a graphite furnace.

    b. A tungsten-halogen background corrector is used for accurate and reproducible background correction during atomization.

Range and Sensitivity

1. The actual range of the procedure has not been established. The lowest concentrations reported are as follows:

0.005 μg/ml Cd,
0.005 μg/ml Cr,
0.005 μg/ml Ni, and
0.01 μg/ml Pb.

2. The working range may be extended by adjusting the aliquots used for the sample and/or for injecting into the graphite furnace.

Interferences

1. Matrix interferences are controlled by using the method of standard additions in making the analysis.

Precision and Accuracy

1. Precision expressed as standard deviation is as follows:

0.5 μg/l for Cd determined at 5 μg/l,
0.8 μg/l for Cr determined at 5 μg/l,
1.2 μg/l for Ni determined at 10 μg/l, and
1.7 μg/l for Pb determined at 10 μg/l.

2. The accuracy of the analytical method has not been established.

Apparatus

1. Atomic absorption spectrophotometer with both a deuterium arc and a tungsten-halogen background corrector and a sample area which will accommodate the graphite furnace.
2. Graphite furnace attachment which is programmable from ambient up to 3,000°C.
3. Graphite tubes, pyrolytically coated.
4. Hollow cathode lamp or electrodeless discharge lamp with power supply for each desired element.
5. Multirange, variable speed recorder.
6. Eppendorff pipettes: 20 µl.
7. pH meter with a combination reference-indicating electrode.
8. Clinical centrifuge.
9. Vortex mixer.
10. Glass centrifuge cones: 15-ml capacity with Teflon-lined plastic screw cap, Kimax 45166 or Corning 8082.
11. Laboratory glassware.

Reagents

All reagents are reagent-grade or ultra-pure and water is distilled then deionized.

1. Nitric acid, concentrated.
2. Hydrochloric acid, 1.0 N: add 3.6 ml of concentrated hydrochloric acid to about 50 ml of water and dilute to 100 ml.
3. Acetic acid, glacial.
4. Ammonium pyrrolidine dithiocarbamate (APDC) solution, 2 g/100 ml: One gram of ammonium pyrrolidine dithiocarbamate (APDC) is dissolved in 50 ml of water, and the solution is extracted twice with 5 ml of MIBK. This solution is prepared each day.
5. Methyl isobutyl ketone (MIBK).
6. Standard Stock Solutions (commercially available solutions may be used).

a. Standard Stock Solution: cadmium, 1,000 μg/ml;

b. Standard Stock Solution: chromium, 1,000 μg/ml;

c. Standard Stock Solution: lead, 1,000 μg/ml; and

d. Standard Stock Solution: nickel, 1,000 μg/ml.

7. Standard working solutions: Make successive dilutions of the stock solutions to obtain a working standard solution for each element with the following concentrations: 0.025, 0.05, and 0.1 μg/ml, respectively. Make the final solution 2% with nitric acid.

Procedure

1. Cleaning of Equipment. All glassware should be thoroughly cleaned and rinsed with hot water, nitric acid (1:1), and distilled-deionized water. This glassware should be used exclusively for heavy metal analysis in urine.

2. Sample Handling and Preservation

   a. Urine samples are made one percent (v/v) acetic acid.

   b. The analysis should be completed immediately following extraction to avoid loss of sensitivity.

3. Sample Preparation

   a. Cadmium, Lead, and Nickel

      (1) Pipet 5 ml of urine which contains 1% acetic acid into each of three 15-ml centrifuge tubes. Label these tubes Nos. 1, 2, and 3, respectively.

      (2) To Tube No. 1, add 1 ml deionized water. To Tube No. 2, add 1 ml of 0.025 μg/ml Cd or 1 ml of 0.05 μg/ml Ni or Pb standards. To Tube No. 3, add 1 ml of 0.05 μg/ml Cd or 1 ml of 0.1 μg/ml Ni or Pb standards. Mix each sample on the Vortex mixer.

      (3) Adjust the pH of each solution to 2.8 ± 0.2 with dropwise addition of 1.0 N HCl. Mix with the Vortex mixer.

      (4) Add 0.2 ml of the APDC solution to each tube and mix thoroughly.

      (5) Pipet 5.0 ml of MIBK into each tube. Cap with the Teflon-lined cap and shake for 2 min.

      (6) Centrifuge (2,800 rpm) for 10 min.

b. Chromium

(1) Pipet 5 ml of urine which contains 1% acetic acid into each of three 15-ml centrifuge tubes. Label these tubes Nos. 1, 2, and 3, respectively.

(2) To Tube No. 1, add 1 ml deionized water. To Tube No. 2, add 1 ml of 0.025 μg/ml Cr standard. To Tube No. 3, add 0.05 μg/ml Cr standard. Mix each sample on the Vortex mixer.

4. Analysis of Sample

a. Cadmium, Lead, and Nickel

(1) Operating the instrument according to the manufacturer's instructions, carefully inject 20 μl of the top layer into the injection port of the graphite furnace. Begin with the unspiked sample and work toward the highest spiked sample.

(2) The following parameters have been used on both a Perkin-Elmer 373 atomic absorption spectrophotometer with a Perkin-Elmer HGA-2100 graphite furnace and with a Perkin-Elmer Model 5000 atomic absorption spectrophotometer with a Perkin-Elmer Model HGA-500 graphite furnace:

Element	Ni	Cd	Pb
Wavelength (nm)	232.0	228.0	283.3
Slit Setting (nm)	0.2	0.7	0.7
Mode	Absorbance	Absorbance	Absorbance
Signal	Peak Height	Peak Height	Peak Height
Background Correction	Deuterium arc	Deuterium arc	Deuterium arc
Purge Gas	Argon	Argon	Argon
Gas Interrput	No	No	Yes
Dry (temp/total time)	130°/20 sec	130°/30 sec	130°/30 sec
Char (temp/total time)	900°/40 sec	350°/30 sec	600°/50 sec
Char Ramp Time	-	10 sec	20 sec
Atomize (temp/ total time)	2,700°/10 sec	2,000°/8 sec	2,300°/10 sec

b. Chromium

(1) Operating the instrument according to the manufacturer's instructions, carefully inject 20 μl of the sample into the injection port of the graphite furnace. Begin with the unspiked sample and work toward the highest spiked sample.

(2) The following parameters have been used on a Perkin-Elmer Model 5000 atomic absorption spectrophotometer with a Perkin-Elmer HGA-500 graphite furance.

Wavelength (nm)	357.9
Slit setting (nm)	0.7
Mode	Absorbance
Signal	Peak Height
Background Correction	Tungsten-Halogen
Purge Gas	Argon
Gas Interrupt	Yes
Dry (temp/total time)	105°/100 sec
Char (temp/total time)	1,300°/240 sec
Char Ramp Time	120 sec
Atomize (temp/total time)	2,700°/10 sec

Calibration

1. Determine the peak height (absorbance) from the display or the chart recorder.
2. Prepare a "Standard Additions" calibration curve by plotting peak height versus concentration of the standards added to the samples.

Calculations

Determine the metal concentration in the sample from the standard additions plot.

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