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Metal Ion Extraction with a Thiol Hydrophilic Resin

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The application of 30% *N,N'*-methylenebis(acrylamide) cross-linked poly[*N*-((acryloylamino)methyl)mercaptoacetamide] resin for the concentration of metal ions from aqueous solutions has been investigated. A flow injection analysis method using a color forming reagent was developed, allowing fast cation assays. The pH dependence of the metal extraction for Na, Ca, Mn(II), Fe(II), Co(II), Ni(II), Cu(II), Zn(II), Ag(I), Cd(II), Hg(II), Pb(II), and UO_2^{2+} was studied. Heavy metals and Cu(II) exhibit a high affinity toward the thiol functions of the resin (half extraction pH < 2), with fast fixation kinetics due to the hydrophilic matrix. The maximum resin capacity depends on the metal ion, due to the formation of ML_2 or/and MLL' complexes involving thiol functions and anion ligands. The resin selectivity determined at pH 5.5 is, in increasing order, Zn(II), Cd(II), Pb(II), Cu(II), and Hg(II).

The study of the removal of metal ions from diluted aqueous solutions, either for pollution control or for raw material recovery, has been taking on increasing importance in recent years. Van Grieken et al. (1) compared various procedures for the concentration of trace metals and concluded that the use of immobilized metal complexing groups are the simplest methods, leading to a good recovery ratio.

However, the most efficient use of chelating resins requires that the following conditions are met: (A) a good ion uptake into the macromolecular network, allowing fast fixation kinetics; (B) a high stability of the matrix and grafting against hydrolysis; (C) a selective ion fixation, i.e., no affinity for alkali or alkaline earth ions, the concentration of which is usually quite high in natural waters; (D) a high strength fixation of the metal ions to be extracted but also (E) a highly reversible complexation, thus allowing a total recovery of ions without resin destruction.

The sulfur-containing chemical groups act as selective ligands with a high bond stability for transition-metal ions (2). Several papers (3-14) describe the immobilization of thiol-containing groups on various more or less hydrophobic matrices, particularly styrene-divinylbenzene copolymers (3-8) or polymethacrylate resins (3, 9-11).

We have prepared a thiol group containing chelating resin, having the structure depicted in Figure 1. The polyacrylamide matrix allows condition (A) to be met owing to the hydrophilicity of the amide group. The thiol function grafting has been achieved through a methylenediamide linkage having a good stability in a wide pH range. We have reported in

another paper (15) the synthesis and some physicochemical properties of this new resin.

A preliminary study of the complexation of various metal ions and of the extractive and recovery properties of the resin are presented here. Moreover, this work entailed the development of a fast spectrophotometric method for the assay of metal ions by flow injection analysis (FIA) (16, 17).

EXPERIMENTAL SECTION

Reagents. All experiments were carried out with a 30% *N,N'*-methylenebis(acrylamide) cross-linked poly[*N*-((acryloylamino)methyl)mercaptoacetamide] resin (Figure 1) prepared according to the previously described procedure (15). Its capacity in thiol groups was $2.80 \pm 0.15 \text{ mmol g}^{-1}$. Metal ion solutions were prepared from analytical grade perchlorate salts (Ventron GmbH, Karlsruhe, West Germany). Suitable concentrations were obtained by dilution of a 10^{-1} M NaClO_4 solution, to maintain a constant ionic strength, except for mercury(II) solutions, where we had to use 10^{-1} M acetate buffer. 4-Pyridylazoresorcinol (PAR) was used as purchased (Prolabo, Paris, France) in 125 mg L^{-1} solutions, prepared as described by Phillips and Fritz (8).

Metal Ion Determination. PAR colorimetry was used when only one metal species was present. Otherwise, differential pulse anodic stripping polarography was chosen.

PAR Colorimetry. We used a FIA type colorimetric method (Figure 2) originally developed for detection after HPLC separation (8, 18).

A LC20 Pye Unicam pump was used to generate a 2 mL min^{-1} PAR dye flow in 0.5 mm i.d. Teflon tubing (18) into which $20 \mu\text{L}$ of metal ion solution samples were injected through a Model 7125 rotary valve (Rheodyne Inc., Berkeley, CA). After mixing in a low-volume ($<150 \mu\text{L}$) magnetically stirred chamber, the differences in coloration are detected in a $10\text{-}\mu\text{L}$ HPLC measure cell of a ISCO UA5 UV/Vis spectrophotometer (Instrumentation Specialties Co., Lincoln, NE) working at 510 nm.

Differential Pulse Anodic Stripping Polarography (DPASP). Hg(II) assays were performed with a Solea PRG5T pulse polarograph connected to a vitreous carbon rotating disk electrode (Solea-EDI, Villeurbanne, France). Zn(II), Cd(II), Pb(II), and Cu(II) assays were performed with Princeton Applied Research equipment consisting of a Model 364 pulse polarograph and a Model 303 hanging mercury drop electrode (EG and G Princeton Applied Research, Princeton, NJ).

In both cases, the following conditions were chosen: differential pulse 50 mV, deposition time 90 s, and anodic scan rate 5 mV s^{-1} .

Metal Ion Uptake as a Function of pH. A batch technique was used, the chelating functions always being in excess to the metal. Dry resin (100 mg) (0.28 mmol thiol functions) was suspended with a 0.1 mmol cation perchlorate solution (100 mL). The pH was adjusted by adding 0.1 M HClO_4 or 0.1 N NaOH . Suspensions were stirred for 1 h and then filtered and the con-

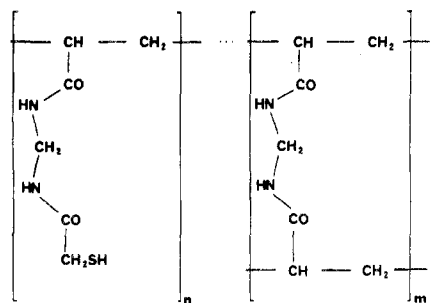


Figure 1. Chemical structure of poly[*N*-((acryloylamino)methyl)mercaptoacetamide] resin.

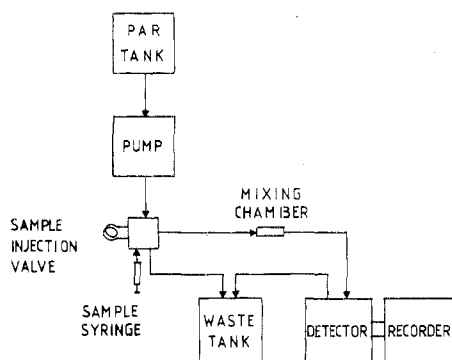


Figure 2. Schematic diagram of the PAR-FIA apparatus.

centrations of metal ions were determined both in the solution and in the resin after acidic regeneration, by PAR FIA.

Resin Regeneration. The resin was suspended for 1 h in an acidic medium and then filtered, the recovered amount of ion being titrated by PAR FIA. According to the fixed ion, the regeneration pH was determined from the results of ion uptake pH dependence.

Metal Uptake as a Function of Time. The following batch method was applied: 100 mg of dry resin was suspended in pH 5.5 water for some minutes to allow the complete swelling of the resin and then 0.1 mmol of pH 5.5 cation perchlorate solution was added, final volume being 100 mL. A glass electrode allowed us to follow the evolution of ion uptake through the increase of H^+ concentration in solution except for Hg(II) where the metal ion concentration was determined by potentiometry (TAT4 Solea) with an amalgamated gold electrode.

Resin Capacity and Sorption Isotherm. The procedure used in the study of uptake variation with pH was applicable, except that pH was kept fixed at 5.5 for the duration of the equilibration and that the metal ion concentration varied for each trial. In the cases of UO_2^{2+} and Hg(II), we had to use 10^{-1} M acetate buffer (pH 5.5) to avoid a precipitation of metal hydroxide.

Resin Selectivity. A similar procedure as above, but with two different 0.1 mmol cations (except Hg(II) 0.2 mmol), was used. Metal ions assays were performed with DPASP.

Resin Stability. The thiol function content in the resin is determined according to previously described procedures (15), before metal ion fixation, after resin regeneration, and after an eventual thiol reduction by mercaptoethanol (15).

RESULTS AND DISCUSSION

Metal Ion Concentration Determination by PAR-FIA.

The application of PAR-FIA allowed us to determine with a good reproducibility (standard deviation less than 5%) and a good sensitivity, the cation concentration in a large series of small samples (20 μ L). The transit time through the apparatus, shorter than 20 s (maximal absorbance at 6 s), allowed an assay rate of better than 60 injections/h.

Figure 3 shows the signals obtained at 510 nm (λ_{max} for the different metal-PAR complexes being from 495 to 530 nm (19)) for samples with an increasing metal ion content. The injection of distilled water induces a high transmittance peak leading to a negative signal due to the dilution of PAR by

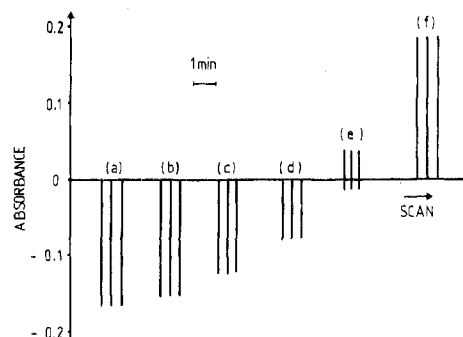


Figure 3. Typical PAR-FIA signals: standard Zn(II) solutions containing (a) 0, (b) 10^{-6} M, (c) 5×10^{-6} M, (d) 10^{-5} M, (e) 2×10^{-5} M, (f) 4×10^{-5} M, (each sample (20 μ L) being injected in triplicate).

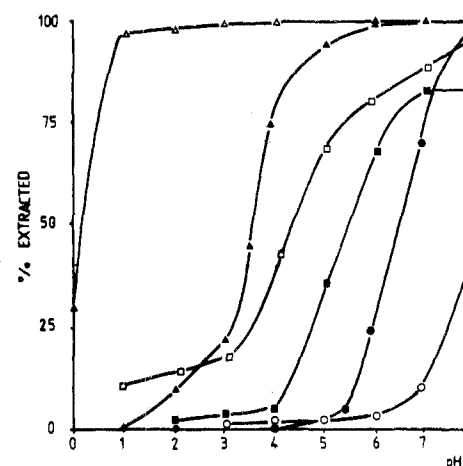
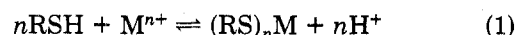


Figure 4. Metal uptake as a function of pH: \circ Mn(II); \bullet Fe(II); \blacksquare Co(II); \square Ni(II); Δ , Cu(II); \blacktriangle , Zn(II).

water. Absorbance of the PAR-metal complex increases with the concentration of metal from a negative (low absorption) to a positive value. However, there is a concentration domain (viz., about 2×10^{-4} M for Zn(II) assay (Figure 3)) in which the accuracy of the measure is lowered: the signal is weak owing to the contrary effects of a positive deviation due to the absorption of the metal-PAR complex and a negative one from PAR dilution, its particular shape leads to difficulties in measurement. In fact, the apparent base line is the 510 nm absorbance of 125 mg L^{-1} PAR (18, 19). To be able to obtain valid calibration curves, we have to start from the water peak, representative of uncomplexed PAR at the experimental, sample induced, concentration. The obtained curves are thus linear in the cation concentration range from 10^{-6} to 10^{-4} M, data reduction being peak height. It should be noted that a calibration may be necessary with each new batch of PAR, and it was systematically done.

All the studied cations can be titrated by this method except Ag(I), the PAR complex of which shows only a slight absorption, its concentration being determined by potentiometry. The signal of Hg(II) is weak but increases with transit time through the apparatus, most particularly for concentrations over 5×10^{-5} M, probably owing to slow complexation kinetics in the experimental conditions (sample-PAR mixing time less than 6 s). Therefore, we have preferred to determine its concentration by a classical PAR spectrophotocolorimetric method.

Metal Ion Uptake as a Function of pH. The metal-thiol complexation releases protons according to the reaction



and thus is pH dependent. The extraction behavior of the resin toward various cations (Na, Ca, Mn(II), Fe(II), Co(II),

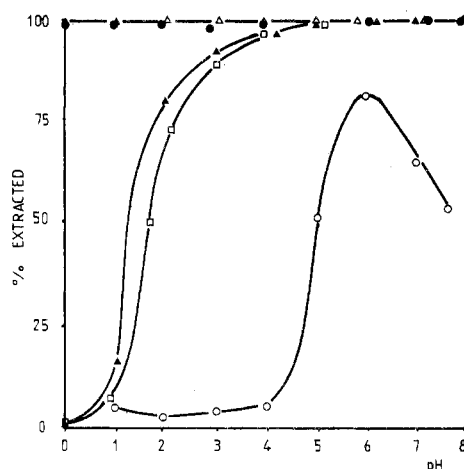


Figure 5. Metal uptake as a function of pH: Δ , Ag(I); \blacktriangle , Cd(II); \bullet , Hg(II); \square , Pb(II); \circ , UO_2^{2+} .

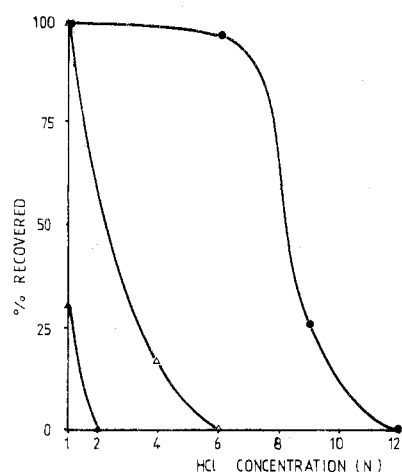


Figure 6. Metal recovery in HCl solutions: \blacktriangle , Cu(II); Δ , Ag(I); \bullet , Hg(II).

Ni(II), Cu(II), Zn(II), Ag(I), Cd(II), Hg(II), Pb(II), U(VI)) was investigated in aqueous solution over the pH range 0–8. The data from these experiments are presented in Figures 4–6.

Over this pH domain, alkali and alkaline earth ions are not extracted and therefore exhibit no affinity for the thiol function. Generally, the uptake of the other cations increases with pH and is quantitative over pH 5 for the heavy metals and Cu(II). However, for the oxygenated cation, UO_2^{2+} , the extraction curve shows a marked fixation decrease over pH 6. This apparent capacity loss may be due to the competitive hydroxide complex formation. In any case, a total recovery of the fixed metal can be achieved by contacting the resin with low pH solutions (Figure 6).

These data (percent extracted vs. pH) can be summarized in terms of half extraction pH ($\text{pH}_{1/2}$) (20). A comparison of $\text{pH}_{1/2}$ values with the available stability constants of corresponding sulfides (21) shows quite a good agreement, and these values lead to the same ordering as does Irving and William's complex stability rule (22) for divalent first row transition metals (Figure 4), i.e., $\text{Mn(II)} < \text{Fe(II)} < \text{Co(II)} < \text{Ni(II)} < \text{Cu(II)} > \text{Zn(II)}$. Therefore, the $\text{pH}_{1/2}$ values can be considered as a good evaluation of the affinity of the metal toward the resin.

On the other hand, there is a wide difference between the $\text{pH}_{1/2}$ values of the different cations (Figures 4 and 5). Thus, the resin can be used for metal ion preconcentration and, furthermore, for selective separation and recovery, according to the equilibration pH.

Ion Uptake as a Function of Time. The knowledge of fixation kinetics is of the utmost importance for an eventual

Table I. Metal Uptake Parameters

	max metal uptake, ^a mg g^{-1}	$[\text{H}^+ \text{ released (mmol} \cdot \text{g}^{-1})] / [\text{metal uptake (mmol} \cdot \text{g}^{-1})]^{\text{a,b}}$	$(\text{dpH}/\text{dt})_i$, s^{-1}
Ca	0		
Mn(II)	8.8	1.9	0.10
Fe(II)	33		0.25
Co(II)	41	2.1	0.80
Ni(II)	67.5	1.9	0.85
Cu(II)	82.5	2.1	0.70
Zn(II)	111	1.6	0.90
Ag(I)	421	0.75	1.05
Cd(II)	146	1.5	1.0
Hg(II)	602		1.9 ^c
Pb(II)	342		1.4
UO_2^{2+}	200		

^a At pH 5.5. ^b Measured at maximum fixation. ^c Inferred from the variation of Hg(II) concentration.

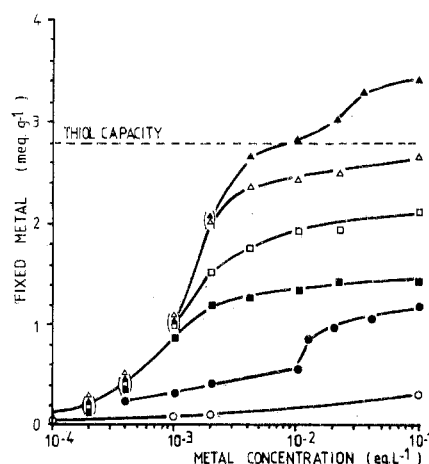


Figure 7. Metal uptake as a function of metal concentration: \circ , Mn(II); \bullet , Fe(II); \blacksquare , Co(II); \square , Ni(II); Δ , Cu(II); \blacktriangle , Zn(II).

use of a resin in a dynamic metal ion extraction process. Its determination is possible through a measure of the variation of pH during metal uptake (reaction 1). However, the reaction rate is itself a function of pH, and only the initial slope of the plot of pH vs. time of contact is indicative of exchange kinetics at the optimal extracting pH (pH 5.5). The chelating functions are always kept in excess to the ions to stay in the range of practical extraction processes.

The results, listed in Table I, show the same kind of variation as does the stability of the metal complex yielded by the reaction, as evaluated from $\text{pH}_{1/2}$, with only Ag(I) and Cu(II) as exceptions. This may be related to redox reactions involving the thiol group with these oxidizing cations, as demonstrated in many cases for Cu(II) (2, 23).

All the cations in this study show high exchange rate, except for Fe(II) and Mn(II), the affinity of which for the thiol group is quite low ($\text{pH}_{1/2} \geq 7$). For instance 90% of Zn(II) in a 5×10^{-3} M solution is extracted in less than 5 min when pH is kept constant at 6 (15). In the same conditions, where hydrophobic matrix resins need equilibration times of some hours (4, 6, 12–14), the studied resin, such as those developed by Slovak (10), reach equilibrium in some minutes probably owing to their hydrophilicity.

Resin Capacity and Sorption Isotherms. Studies of the sorption isotherms of the resin have been carried out to determine the behavior of its chelating sites during the saturation and its maximal capacity (Table I). The results are plotted in Figures 7 and 8.

The comparison between the amount of the fixed metal ion and the number of released protons during the uptake, leads

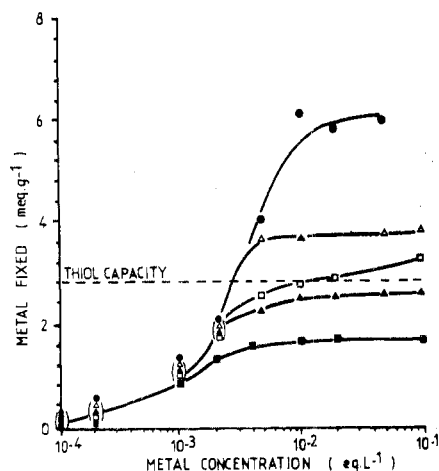


Figure 8. Metal uptake as a function of metal concentration: Δ , Ag(I); \blacktriangle , Cd(II); \bullet , Hg(II); \square , Pb(II); \circ , UO_2^{2+} .

to distinguish different classes of cations. For Mn(II), Co(II), and Ni(II) (first cation class), two protons are liberated as each divalent ion is chelated (Table I). This is in good agreement with a ML_2 chelate formation according to reaction 1.

On the contrary, the number of released protons is significantly inferior to the expected value in the case of Zn(II) and Cd(II), suggesting a different fixation scheme. On the other hand, Hg(II), Pb(II) and Zn(II) exhibit an uptake (Figures 7 and 8) which exceeds the resin capacity as determined by reaction 1. In the case of Hg(II), a mixed ligand MLL' may be formed, the number of Hg(II) fixed being equal to the thiol content (stoichiometry 1:1) (Figure 8). This kind of complex has been mentioned with chloride ions as second ligand (2, 5). We suggest a reaction scheme



(in this study, ligand A is acetate). In a previous study (15), we have reported for Zn(II) the possibility of the existence of both RSZn^+ and $(\text{RS})_2\text{Zn}$. These complexes may be present for similar metal ions, as Pb(II), leading to a resin capacity higher than the first cation class one.

In the case of Cu(II), redox reactions can occur simultaneously with chelation, leading to the formation of Cu(I) and disulfide groups, as demonstrated by previous studies with thiol compounds (2, 23). The existence of redox reactions is evidenced by the important decrease of the thiol content after metal release (Table III). It is likely that complexes between the Cu(II) and Cu(I) ions and the thiol and disulfides ligands take part in the fixation mechanism although the proton release (Table I) would be in favor of the ML_2 chelate formation.

For Ag(I), one proton is liberated as each 1.35 Ag(I) ions are fixed (Table I). Likewise, the ratio of Ag(I) capacity to thiol function resin content is 1.35. These results may be interpreted by the simultaneous occurrence of Ag_2SR^+ and AgSR complexes (24) in the 1:2 ratio.

Resin Selectivity. The ions in natural or waste waters constitute generally a complex mixture, only some of which are to be extracted and/or recovered by the ion exchange process. It is therefore important to obtain knowledge about the interaction of different ions in a solution on the uptake data of a particular metal.

In this aim, competitive extractions have been carried out for pairs of metals, the concentrations of each not being enough to saturate the resin, but with a total concentration in excess to the metal capacity. The results are presented in Table II. Hg(II) is extracted preferentially to any other studied metal and the order of selectivity is the same as that of affinity deduced from $\text{pH}_{1/2}$, i.e., by increasing order: Zn(II),

Table II. Competitive Uptake of Metal Pairs at pH 5.5

metal pair A/B	initial concn A/B	final concn A/B	log of the apparent exchange constant $\log K_A^B$
Zn	1	18.5	2.34
Cd	1	10.5	2.38
Zn	0.5	195	<i>a</i>
Hg	1	1.35	0.25
Cd	1	41	2.34
Cu	0.5	47.5	<i>a</i>
Pb	1	43.5	2.52
Hg	0.5	87	<i>a</i>
Cu	0.5	2.35	<i>a</i>

^a Not calculated because of the formation of mixed ligand MLL' complexes.

Table III. Variation of Thiol Capacity with Metal Uptaken (Initial Resin Capacity 2.80 ± 0.15)

metal	thiol capacity after	
	metal release, mmol g ⁻¹	mercapto-ethanol reduction, mmol g ⁻¹
Ca	2.6	2.8
Mn(II)	2.7	2.8
Fe(II)	1.0	2.5
Co(II)	1.1	2.1
Ni(II)	1.5	2.4
Cu(II)	0.5	2.6
Zn(II)	2.5	2.6
Ag(I)	1.4	2.1
Cd(II)	2.9	
Hg(II)	1.7	2.7
Pb(II)	2.8	
U(VI)	2.9	
blank after 24 h in 12 N HCl	1.9	2.4

Cd(II), Pb(II), Cu(II), and Hg(II).

From the results in Table II, apparent exchange constants can be calculated for pH 5.5 and there is quite a good agreement between the values from different exchange data.

Resin Stability. Dry resin can be left standing for some months, in closed containers, without any loss in capacity. Nevertheless, the capacity of the resin may vary during the metal ion fixation, due to the oxidizability of the thiol function. This phenomenon was investigated through a comparison of the total capacity after acid regeneration and after reduction of the eventually formed disulfide groups, by mercaptoethanol. The results are listed in Table III.

Ca, Mn(II), Zn(II), Cd(II), Pb(II), and UO_2^{2+} can be considered as oxidatively inert toward the resin. The other cations generally show an oxidation effect, either by themselves as Cu(II), Ag(I), and Hg(II) or by acting as catalyst for the thiol oxidation by O_2 (23). For the latter, the original thiol capacity is harder to recover. In addition, the capacity loss recorded during the fixation of Ag(I) or Hg(II) is also related in part

to the regeneration in concentrated (12 N) hydrochloric acid, as can be seen by contacting the resin during 24 h with such a solution, inducing a 15% thiol capacity loss (Table III). However, except for Ag(I) and Co(II), the recovery of the initial thiol capacity is complete or nearly complete.

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Determination of Iodoamino Acids and Thyroid Hormones in a Urine Matrix by Neutron Activation Analysis

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By combining neutron activation detection with gel filtration and high-performance liquid chromatography (HPLC) employing C_8 and C_{18} reversed-phase columns, it is possible to detect and quantitate in a urine matrix trace quantities of 3-iodotyrosine, 3,5-diiodotyrosine, 3,5-diiodothyronine, 3,5,3'-triiodothyronine, and 3,5,3',5'-tetraiodothyronine. Neutron activation of HPLC fractions where various thyroid derivatives elute is highly specific and sensitive for the ^{128}I activity. Experiments with spiked standards in urine show a quantitative response with a detection limit in the nanogram range utilizing a nuclear reactor with a power level greater than 1×10^{12} neutrons $\text{cm}^{-2} \text{ s}^{-1}$.

The determination of total and hormonal iodine in serum and urine is important for the study of thyroid physiology in health and disease. The two main techniques currently used for total iodine are colorimetric procedures (1, 2) [involving oxidation-reduction catalysis of the As(III)-Ce(IV) system] and neutron activation analysis (NAA) (3-6) involving multiple wet-chemical pre- or postirradiation operations such as oxidation and reduction, solvent extraction, use of anion resins, distillation, and/or precipitation. Currently there is more interest in the determination of iodoamino acids and hormonal iodine in various biological matrices. Modified radioimmunoassay (RIA) procedures have been employed for the various routine clinical and research determinations (7-9) of 3,5-diiodothyronine (T_2), 3,5,3'-triiodothyronine (T_3), and

3,5,3',5'-tetraiodothyronine (T_4). There has been activity in recent years in the development of high-performance liquid chromatography (HPLC) and gas chromatography (GC) procedures for iodoamino acid and hormonal iodine determinations in water, serum, thyroid tissue, and thyroglobulin (10-17).

Trace iodine exists in urine as >80% iodide ion and as <20% organic compounds (18). The total iodine concentration in clinical urine samples can vary from $\sim 0.1 \mu\text{g/mL}$ to over $2000 \mu\text{g/mL}$ with a mean concentration of approximately $0.2 \mu\text{g/mL}$ urine. While there have been various colorimetric (1, 2) and neutron activation analysis (3-6) procedures for total iodine determinations in urine, there are no NAA techniques for determining iodoamino acids and hormonal iodine. One characteristic of the thyroid hormones T_2 , T_3 , and T_4 , and their precursors MIT and DIT is that they contain ^{127}I , the only stable isotope of iodine. Neutron irradiation of ^{127}I produces ^{128}I ($T_{1/2} = 25.2 \text{ min}$) by radiative neutron capture in a nuclear reactor. Not only is urine a complex biological matrix for analysis because of the great number of biological and organic compounds present but it contains a large quantity of the radioactivatable elements sodium and chlorine. For any NAA procedure for trace elements, it is important to remove either the trace element from the sodium and chloride ions or the sodium and chloride ions from the biological matrix. It was the intent of this study to combine NAA with HPLC for the determination of iodine-containing thyroid compounds in urine. Most HPLC detectors (UV-VIS, fluorescent, and electrochemical) are sensitive only for a select group of com-