CHEMISTRY LETTERS, pp. 363-366, 1981.

# THIN-LAYER CHROMATOGRAPHY AND QUANTITATIVE PHOTOACOUSTIC SPECTROMETRY. APPLICATION TO THE DETERMINATION OF NANOGRAM AMOUNTS OF COBALT, COPPER, IRON, AND NICKEL AS THEIR CHELATES WITH 1-(2-PYRIDYLAZO)-2-NAPHTHOL

Shigero IKEDA, Yoshiki MURAKAMI, and Kunihiko AKATSUKA†
Department of Chemistry, Faculty of Science, Osaka University,
Toyonaka, Osaka 560
† Present address: Kitami Institute of Technology, Kitami 090

A method is described for the determination of metal chelates on thin-layer chromatography plates using photoacoustic spectrometer. Nanogram amounts of Co, Cu, Fe and Ni can be determined up to (40-50) ng, respectively, with less than 5% of c.v. values. The detection limit is much improved by this method, e.g. 0.1 ng.

Thin-layer chromatography (TLC) is fast, simple and highly effective technique for the separation of mixtures, so the separation of the metal chelates by TLC seems to be very attractive, particularly for simultaneous trace determination of several metal ions. For the analysis of the compounds separated by TLC plates, optical spectrometry and reflectance spectrometry are of general use. However, these methods occasionally encountered difficulties for the direct quantitative determination of the species on the TLC plates, because of opacity and light-scattering properties of the TLC adsorbent. Photoacoustic spectrometry (PAS) is a new analytical technique for obtaining the thermal emission spectra of solids following the optical light absorption , and offered the direct measurements of chemical species on TLC plates. The spectra are quite coordinated to that of the light absorption spectra in case of the absence of decay by the fluorescence emission process. Rosencwaig et al.[1)] have shown the applicability of PAS for TLC analysis, and Castleden et al.[2)] tried the quantitative determination of fluorescein on TLC plates in the range 0.2-2 μg. No investigations on the quantitative determination of metal compounds by TLC and PAS have been attempted so far. This paper reports the application of PAS in the quantitative determination of nanogram amounts of Co, Cu, Fe and Ni as 1-(2-Pyridylazo)-2-naphthol (PAN) chelates which were separated each other by TLC after solvent extraction.

All four metal ions (Co,Cu,Fe and Ni) were extracted into chloroform from aqueous solution at pH 5-6 in the presence of 0.1M potassium bromide using ethanolic PAN solution.[3)] The chloroform solution containing metal-PAN chelates was evaporated to dryness and then the residue dissolved in 0.1-0.5 ml of chloroform if preconcentration was required. A (1-10)μl portion of the resulting solution was spotted on the 10×5 cm aluminum backed silica gel plates (Merck, type 5553, 0.2mm thick of adsorbent) with a disposable pipet (Drummond, microcaps). The plates were previously dried for 30 minutes in the oven at 75°C. Development of the chromatogram was carried out with a solvent mixture dichloromethane - ethanol - acetic acid (100:5:0.1) for 20 minutes. The addition of acetic acid is quite effective for the improvement of the separation of four ions.4) After development of chromatograms, the TLC plates were provided for the quantitative determination by cutting out 7×7 mm squares containing the spots. The square was then placed directly into the PAS cell.

Fig. 1 shows the block diagram of the PAS employed in this study. A 300W xenon arc lamp (Varian, type VIX-300 UV) was used as a light source. A microphone settling carbon black was used in the reference channel of the spectrometer to correct for the fluctuation and vibration of output of the lamp with time and wavelength. All the spectra were recorded in normalized signal against a carbon black standard at 45Hz at a scanning rate of 25 nm/min. The PAS cell and others have been described elsewhere 5)-7); the sample cell was placed inside a foam rubber (20 mm thick) lined cabinet to reduce a noise from floor and surroundings.

Figs. 2 and 3 show the PAS spectra obtained for PAN and its chelates with Co,Cu, Fe and Ni containing 10 ng of each metal examined under the conditions described above excluding Co, and the corresponding spectrum for the silica gel substrate. The PAS spectra quite correspond to the light absorption spectra of the corresponding chelate species extracted into chloroform solution, and can be obtained in the presence of only several nanogram amounts of metals, whereas the solution spectra require the presence of microgram order of metals.8) The calibration curves obtained by plotting the normalized signal intensity against each metal-PAN chelate were linear up to

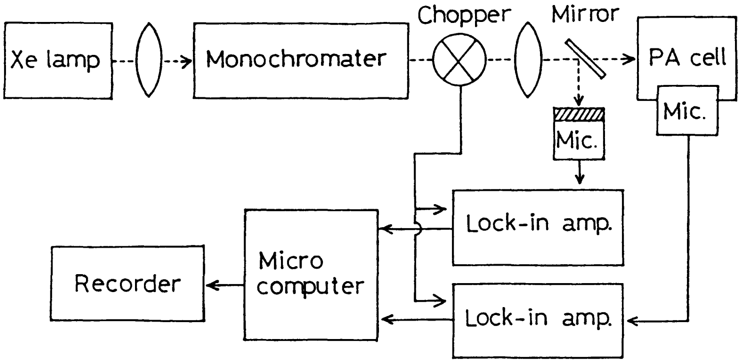
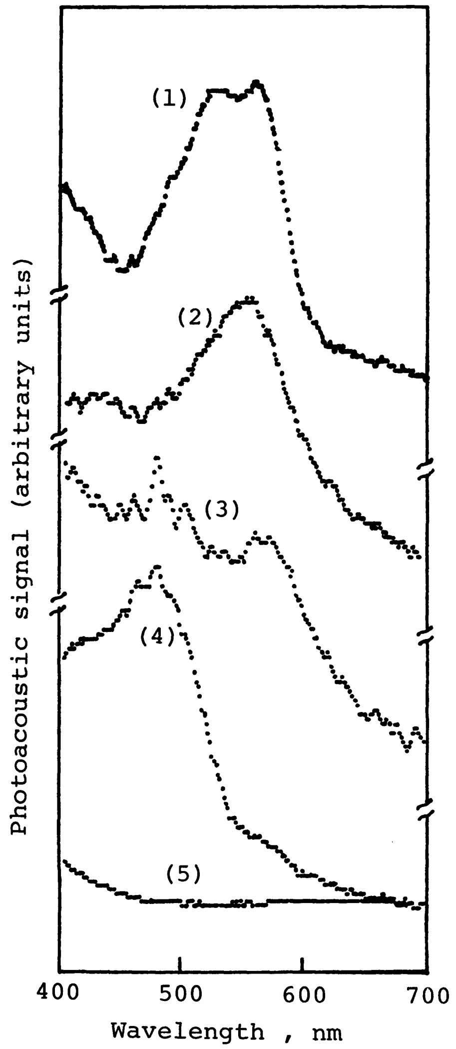


Fig. 1 Block diagram of the P.A. spectrometer.

(40 - 50) ng, respectively, and passed through the origin with adopting the calculated correction intensity by subtracting the PAS intensity from the corresponding intensity of silica gel substrate, i.e. background intensity, as shown in Fig.4. The background intensity of silica substrate is several-hold larger than the calculated correction intensity of 2 ng of cobalt as shown in Figs. 3 and 4. In the determination of trace amounts of metal-chelates,the majorities of the errors are caused by the difference of TLC square size and exposed area of backed-aluminum plate as reported by Castleden et al.[2) This forced them to the isolation of TLC spot from the substrate using a vacuum collection. However, in the present method, the reproducible TLC square size was obtained and no difficulties were found for the subtraction of the background signals.



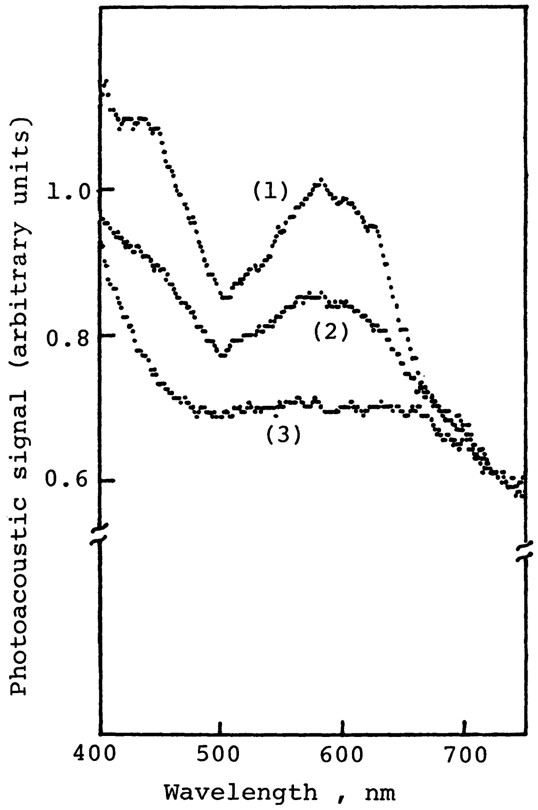


Fig. 3 Photoacoustic spectra of Co-PAN chelates on TLC plates containing (1) 4 ng and (2) 2 ng of cobalt, and (3) the corresponding spectrum of the silica gel substrate.

Fig. 2 Photoacoustic spectra of (4) PAN and of its chelates with (1) Ni, (2) Cu and (3) Fe on TLC plates, and (5) the corresponding spectrum of the silica gel substrate.

The coefficients of variation of measurements in one square have been found to be in the range 0.1 - 0.5%, and that in the plate-to-plate is less than 5% (7 determinations) for samples containing more than 5 ng of metals. The TLC detection limit is much improved by this method, e.g. 0.1 ng; more sensitive value of that has been reported by Galik et al.[3] using reflectance filter photometer, 7-16 ng. If the background correction of the TLC signal would be much improved, the value could be extended to less trace amounts. We are now trying to obtain significant PAS data on that point, and the determination of sub-nanogram, or picogram of metals is now in progress.

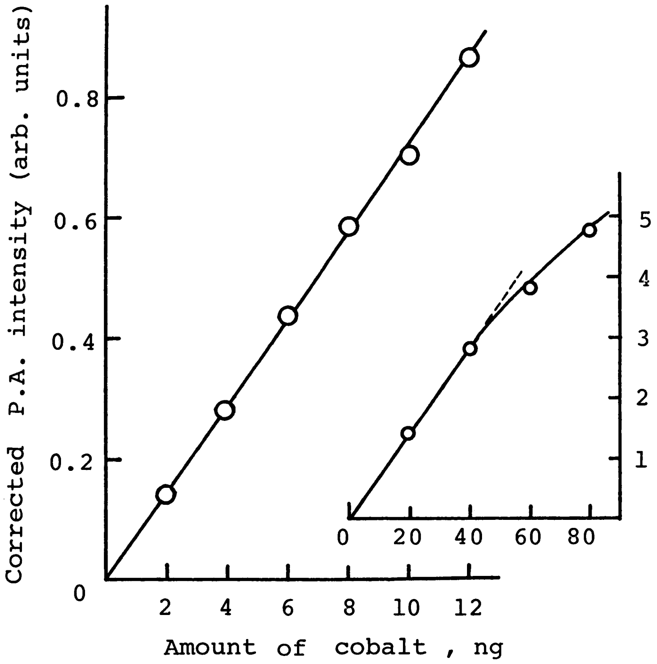


Fig. 4 Calibration curve for cobalt at 570 nm.

## References and Note

1) A. Rosencwaig and S.S.Hall, Anal. Chem., 47 , 548 (1975).
2) S.L.Castleden, C.M.Elliott, G.F.Kirkbright, and D.E.M.Spillane, Anal. Chem., 51 , 2152 (1979).
3) A.Galik and A.Vincourova, Anal. Chim. Acta, 46 , 113 (1963).
4) Unpublished data. $R_f$ values of PAN and of its chelates with Co, Cu, Fe and Ni were $0.6_3$, 0.04, $0.2_0$, $0.9_0$ and $0.7_2$, respectively, under the recommended conditions.
5) Y.Yokoyama and N.Nakamura, Bunseki Kagaku, 28 , 97 (1979).
6) S.Kawamoto, Y.Yokoyama, and S.Ikeda, Bunseki Kagaku, 28 , 142 (1979).
7) S.Kawamoto, Y.Yokoyama, and S.Ikeda, Bull. Chem. Soc. Jpn., 53 , 391 (1980).
8) G.Goldstein, D.L.Manning, and O.Menis, Anal. Chem., 31 , 192 (1959).

(Received January 5, 1981)