glucosaminic acid in copper complex formation, its titration curves in the presence and in the absence of cupric ion were compared with those of glycine under the same conditions. Fig. 4 shows that the titration curves of both amino acids are similar except in the alkaline region of their copper complex where pH depression occurs only with glucosaminic acid. Moreover, a precipitation of hydroxide or hydrolysis products of complex is observed only in the glycine solution.

These results suggest the contribution of OH groups of glucosaminic acid in alkaline regions to the complex formation by their dissociation and coordination to copper.

The authors wish to express their deep gratitude to Prof. Emeritus M. Ishidate of the University of Tokyo for his encouragements and helpful advices. They are also grateful to Mr. Shi-Tsu-Won for his cooperation, and to all the staffs of the central analysis laboratory of this faculty for elemental analysis.

Summary

Stability constants, log K_1 and log K_2, of complexes of D-glucosaminic acid with Cu^{2+}, Ni^{2+}, Pb^{2+}, Zn^{2+}, Co^{2+}, Cd^{2+}, and Mn^{2+} were determined by pH titration method. Copper complexes were found to be the most stable and the contribution of OH groups of glucosaminic acid to the complex formation was observed.

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19. Motoichi Miyazaki, Toshio Imanari, Tamiko Kunugi, and Zenzo Tamura: Gas Chromatography of Copper (II) and Nickel (II) Chelates of Some β-Ketoimine Derivatives of 2,4-Pentanedione and Salicylaldehyde.[*1]

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Recently, gas chromatography has been applied to various volatile metal chelates of 2,4-pentanedione and of its fluoro derivatives, and many investigators[1~10] have suggested that gas chromatography would be very useful for the separation of metal chelates or the micro analysis of metals. However, the study of chelates of other types of ligand than 2,4-pentanedione and its fluoro-derivatives have not been reported.

[*1] A preliminary report was presented at the 84th Annual Meeting of Pharmaceutical Society of Japan in Tokyo (April, 1964).

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1) W.J. Biermann, H. Gesser: Anal. Chem., **32**, 1525 (1960).
2) R.E. Sievers, R.W. Moshier, M.L. Morris: Inorg. Chem., **1**, 966 (1962).
3) R.E. Sievers, B.W. Ponder, M.L. Morris, R.W. Moshier: *Ibid.*, **2**, 693 (1963).
4) W.D. Ross: Anal. Chem., **35**, 1596 (1963).
5) W.D. Ross, G. Wheeler: *Ibid.*, **36**, 266 (1964).
6) R.D. Hill, H. Gesser: J. Gas. Chrom., October 11 (1963).
7) R.S. Juvet, R.P. Durbin: *Ibid.*, December 14 (1963).
8) K. Yamakawa, K. Tanikawa, K. Arakawa: This Bulletin, **11**, 1405 (1963).
9) T. Fujinaga, T. Kuwamoto, Y. Ono: Japan Analyst, **12**, 1199 (1963).
10) J.E. Schwarberg, R.W. Moshier, J.H. Walsh: Talanta, **11**, 1213 (1964).

In this paper, the authors intend to report some observations on the gas chromatography of copper (II) and nickel (II) chelates of β-ketoimine derivatives of 2,4-pentanedione and salicylaldehyde.

Experimental

Materials——All of the metal chelates used are shown in Table I. The ligand compounds and their metal chelates were prepared by the methods given in the literature cited in Table I or by the analogous methods.

Apparatus and Procedure——Shimadzu Gas Chromatograph Model GC-1B (dual column type) equipped with thermal conductivity cell detector was used. A stainless steel tube of U type (1.5 m. length) was packed with 0.5~1.5 % silicon polymer or Apiezon grease on celite or glass beads (60~80 mesh). Glass beads were obtained from Shibata Chemicals, washed with acid and siliconized. The column temperature was 150~220°, and the temperature of sample heater was usually kept 10~40° higher than that. Helium was used as a carrier gas at a flow rate of 40~100 ml./min., and the filament current was 140~160 mA. The chelate compounds were dissolved in $CHCl_3$ and injected to the sample heater. The peak fractions were collected by the ordinary method and were destructed with conc. HNO_3 and 30% H_2O_2. The presence of metal was examined by spot test analysis using sodium rubeanate or dimethylglyoxime.

A Koken DS-402G Spectrophotometer was used for the measurements of IR spectra of metal chelates before and after chromatography. A micro KBr-disk method was used for identification.

Results and Discussion

As a preliminary test, the possibility of application of gas chromatography was examined. As shown in Fig. 1, I gave a definite peak and its shape was very resemble to that of copper chelate of 2,4-pentanedione. The compound II gave a similar chromatogram. As to the copper chelates of N-alkyl derivatives of salicylaldimines,

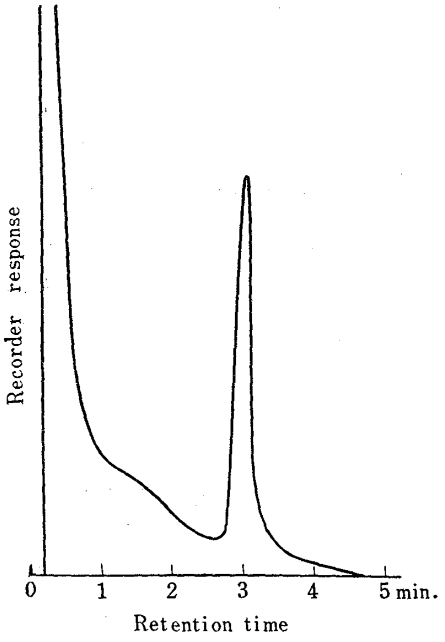

Fig. 1. Gas Chromatogram of Bis-(4-imino-2-pentanono)-copper

Column : 1% Apiezon L (glass beads), 1.5 m.×4 mm.
Temperature : column, 200°
sample heater, 220°
detector, 250°
Carrier gas : He 70 ml./min.

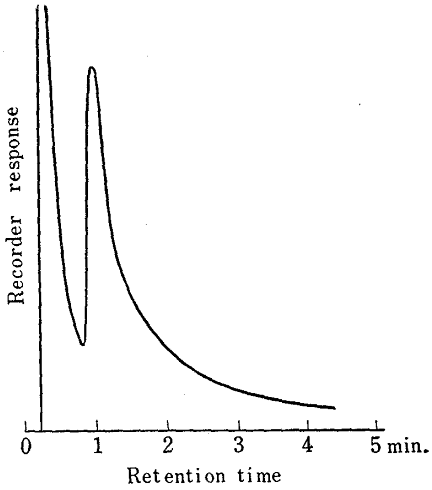

Fig. 2. Gas Chromatogram of Bis-(N-methylsalicylaldimine)-copper

Column : 1.5% SE 30 (Chromosorb W), 1.5 m.×4 mm.
Temperature : column, 160°
sample heater, 200°
detector, 230°
Carrier gas : He 85 ml./min.

III gave also a definite peak as shown in Fig. 2. The gas chromatographic behaviors of IV and V were similar to that of III. Nickel chelate VI showed a very clear gas chromatogram. The result is given in Fig. 3.

Although definite peaks were observed on the gas chromatography of these metal chelates, some elucidations might be necessary to attribute those peaks to the metal chelates. The eluated fractions corresponding to the peaks were collected and the presence of metal was examined by spot test analysis. As shown in Table I, the metals were detected in all of the collected samples. Furthermore, I, II, and VI were found to be thermally stable but III, IV, and V were perceived to be slightly unstable from their infrared spectra. For a typical example, the infrared spectra of VI are given in Fig. 4.

As I and VI gave clear and definite peaks and their retention times are considerably different, a separation of these chelates was examined and a successful result was obtained as shown in Fig. 5.

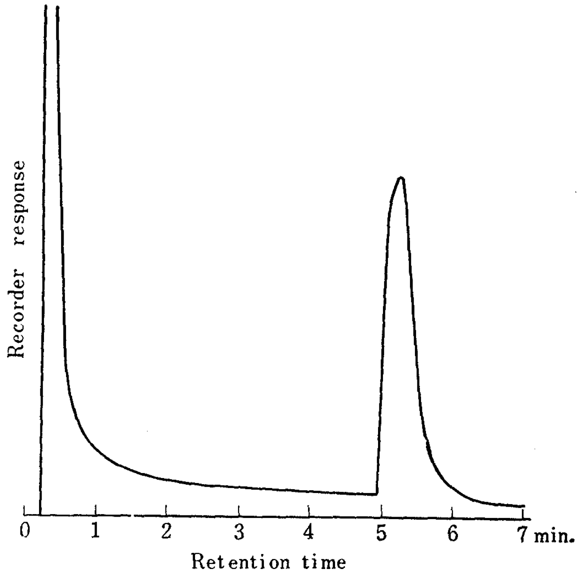

Fig. 3. Gas Chromatogram of Bis-(4-imino-2-pentanono)-nickel

Column : 1% Apiezon L (glass beads), 1.5 m. × 4 mm.
Temperature : column, 200°
sample heater, 220°
detector, 240°
Carrier gas : He 40 ml./min.

Table I. Metal Chelates and Their Gas Chromatographic Stability

No.	Metal chelate	m.p. (°C)	Formula	N (%) Calcd.	N (%) Found	Metal (%) Calcd.	Metal (%) Found[d)]	Column temp. (°C)	M[e)]	Ref.
I	Bis(4-imino-2-pentanono)-copper (II)	185~186	$C_{10}H_{16}O_2N_2Cu$	10.78	10.84	24.80	24.46	180	+	a)
II	Bis(4-methylimino-2-pentanono)-copper (II)	sub. p 120	$C_{12}H_{20}O_2N_2Cu$	9.73	9.90	22.40	23.94	180	+	a)
III	Bis(N-methylsalicylaldimine)-copper (II)	158	$C_{16}H_{16}O_2N_2Cu$	8.45	8.35	19.16	18.59	160	+	b)
IV	Bis(N-ethylsalicylaldimine)-copper (II)		$C_{18}H_{20}O_2N_2Cu$	7.78	7.35	17.65	17.35	150	+~−	b')
V	Bis(N-*n*-propylsalicylaldimine)-copper (II)		$C_{20}H_{24}O_2N_2Cu$	7.22	6.92	16.38	15.63	150	+	b')
VI	Bis(4-imino-2-pentanono)-nickel (II)	246	$C_{10}H_{16}O_2N_2Ni$	10.99	11.23	23.02	23.04	190	+	c)

a) H.F. Holtzclaw, J.P. Collman, R.M. Alire : J. Am. Chem. Soc., **80**, 1100 (1958).
b) P. Pfeiffer, H. Glaser : J. prakt. Chem., **153**, 265 (1939).
b') The analogous method to *b*).
c) R.D. Archer : Inorg. Chem., **2**, 292 (1963); Tong-Ming Hseu, D.F. Martin, T. Mellor : *ibid.*, **2**, 587 (1963).
d) Contents of metal were measured by EDTA titration. No. II compound gave rather high value indicating that the purification was not complete.
e) Detection of metal in peak.

Moreover, the possibility of determination of VI was also investigated. As shown in Fig. 6, a linear relationship exists between the amount of the nickel chelate injected and the peak area or the peak height of the gas chromatogram.

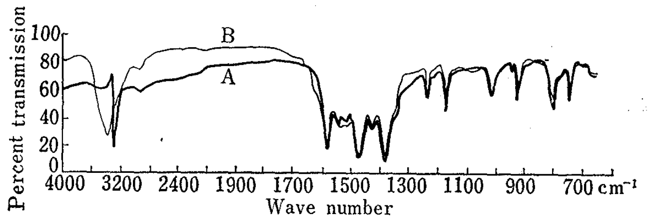

Fig. 4. Infrared Spectra of Bis(4-imino-2-pentanono)-nickel

A : before chromatography
B : after chromatography
Column : 1% SE 30 (anakrom), 1.5 m. × 4 mm.
Temperature : column, 210°; sample heater, 220°; detector, 250°
Carrier gas : He 100 ml./min.

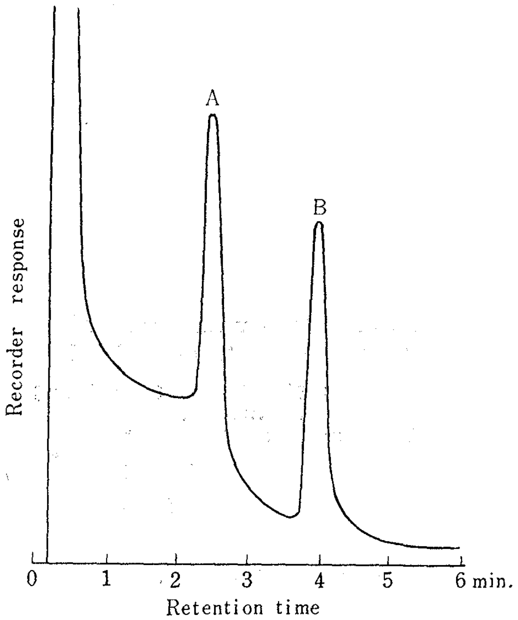

Fig. 5. Gas Chromatogram of the Mixture of Bis(4-imino-2-pentanono)-copper and Bis(4-imino-2-pentanono)-nickel

Column : 0.5% QF-1 (glass beads), 1.5 m. × 4 mm.
Temperature : column, 180°
sample heater, 210°
detector, 240°
Carrier gas : He 45 ml./min.
A : copper chelate
B : nickel chelate

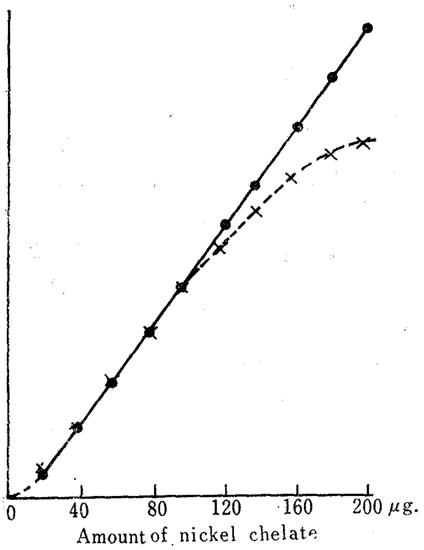

Fig. 6. Calibration Curves of Bis-(4-imino-2-pentanono)-nickel

Column : 1% SE 30 (anakrom), 1.5 m. × 4 mm.
Temperature : column, 195°
sample heater, 220°
detector, 240°
Carrier gas : He 70 ml./min.
• peak area
× peak height

The authors are indebted to Dr. T. Nambara, Associate Professor of this faculty for his pertinent advices and they are also grateful to the staffs of central analytical laboratory of this faculty for elemental analysis and IR spectral measurements.

Summary

Gas chromatography of copper (II) and nickel (II) chelates of β-ketoimine type derivatives of 2,4-pentanedione and salicylaldehyde were investigated. Copper and nickel chelates of 4-imino-2-pentanone gave definite peaks and their separation was attained.

Furthermore, the determination of nickel chelate of 4-imino-2-pentanone was investigated.

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