

Table 1. STABILITY CONSTANTS OF SOME METAL COMPLEXES, IN AQUEOUS SOLUTION AT 20° C.

Unless otherwise indicated, ligand concentrations were 0.05 M and metal ion concentrations were 0.005 M

Cation	Ligand	Log $K_1$	Log $K_2$
Mn <sup>++</sup>	Salicylic acid	5.90	3.9 <sup>a</sup>
Fe <sup>++</sup>	Salicylic acid	6.55 <sup>b</sup>	4.7 <sup>a</sup>
Co <sup>++</sup>	Salicylic acid	6.75	4.7 <sup>a</sup>
Ni <sup>++</sup>	Salicylic acid	6.95	4.9 <sup>a</sup>
Cu <sup>++</sup>	Salicylic acid	10.90	7.85
Zn <sup>++</sup>	Salicylic acid	6.85	pptn.
Cd <sup>++</sup>	Salicylic acid	5.55	pptn.
Fe <sup>+++</sup> (0.001 M)	Salicylic acid (0.01 M)	16.35	11.90
Mn <sup>++</sup>	5-Sulphosalicylic acid	5.10	2.90
Fe <sup>++</sup>	5-Sulphosalicylic acid	5.90	< 4.0
Co <sup>++</sup>	5-Sulphosalicylic acid	6.00	3.60
Ni <sup>++</sup>	5-Sulphosalicylic acid	6.30	3.90
Cu <sup>++</sup>	5-Sulphosalicylic acid	9.50	6.80
Zn <sup>++</sup>	5-Sulphosalicylic acid	6.05	< 4.6
Cd <sup>++</sup>	5-Sulphosalicylic acid	4.85	pptn.
Fe <sup>+++</sup> (0.001 M)	5-Sulphosalicylic acid (0.01 M)	14.60	10.55
Cu <sup>++</sup> (0.0025 M)	2-Hydroxyacetophenone (0.005 M)	6.75	5.70
Cu <sup>++</sup> (0.0025 M)	Methyl salicylate (0.003 M)	5.90	
Cu <sup>++</sup>	Salicylaldehyde (0.02 M)	5.75	pptn.
Cu <sup>++</sup> (0.0025 M)	Salicylamide (0.005 M)	-3.40 <sup>c</sup>	-4.50 <sup>d</sup>
Cu <sup>++</sup>	Catechol	1.25 <sup>c</sup>	0.65 <sup>d</sup>
Fe <sup>++</sup>	Glycine (for comparison)	4.3 <sup>a</sup>	3.5 <sup>a</sup>
Cu <sup>++</sup>	Glycine (for comparison)	3.5 <sup>a</sup>	6.9 <sup>a</sup>
Fe <sup>+++</sup>	Glycine (for comparison)	10.0 <sup>a</sup>	
Fe <sup>++</sup>	Oxine (for comparison)	8.0 <sup>a</sup>	7.0 <sup>a</sup>
Cu <sup>++</sup>	Oxine (for comparison)	12.2 <sup>a</sup>	11.2 <sup>a</sup>
Fe <sup>+++</sup>	Oxine (for comparison)	12.3 <sup>a</sup>	11.3 <sup>a</sup>

(a) Obtained at ligand concentration 0.15 M.

(b) In 0.1 M KCl.

(c) For  $\text{Cu}^{++} + \text{HL} \rightleftharpoons \text{CuL} + \text{H}^+$ .(d) For  $\text{CuL} + \text{HL} \rightleftharpoons \text{CuL}_2 + \text{H}^+$ .

## Ionization Constants for Ionic Strengths of 0.10-0.15

Ligand	$pK$
Salicylic acid	2.93, 13.61
5-Sulphosalicylic acid	2.62, 11.95
2-Hydroxyacetophenone	10.06
Methyl salicylate	9.87
Salicylaldehyde	8.34
Catechol	9.43, ~ 13
Glycine ( $I = 0.01$ ) (ref. 6)	2.24, 9.85
Oxine ( $I = 0.01$ ) (ref. 7)	5.13, 9.89

Table 2. DISTRIBUTION OF CATIONS AMONG LIGANDS, IN NEUTRAL SOLUTION

	Salicylic acid	5-Sulphosalicylic acid	Glycine	Oxine
Cu <sup>++</sup>	1	3.5	45	200,000
Fe <sup>++</sup>	1	10	30	150,000
Fe <sup>+++</sup>	400	300	1	200

salicylic acid, glycine and oxine (8-hydroxyquinoline). Glycine is a typical amino-acid with which salicylic acid and its derivatives would have to compete for metals in the mammalian body, while oxine resembles salicylic acid in being a powerful fungicide.

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<sup>1</sup> Bjerrum, J., Schwarzenbach, G., and Sillén, L. G., "Stability Constants. Part 1: Organic Ligands" (The Chemical Society, Spec. Pub. No. 6, London, 1957).

<sup>2</sup> Bjerrum, J., "Metal Ammine Formation in Aqueous Solution" (P. Haase and Son, Copenhagen, 1941).

<sup>3</sup> Albert, A., *Biochem. J.*, **47**, 531 (1950).

<sup>4</sup> Perrin, D. D., *J. Chem. Soc.* (in the press).

<sup>5</sup> Harned, H. S., and Hecker, J. C., *J. Amer. Chem. Soc.*, **55**, 4833 (1933).

<sup>6</sup> Albert, A., *Biochem. J.*, **54**, 646 (1953).

<sup>7</sup> Albert, A., and Hampton, A., *J. Chem. Soc.*, 505 (1954).

<sup>8</sup> Babko, A. K., *J. Gen. Chem. U.S.S.R.*, **17**, 443 (1947).

<sup>9</sup> Agren, A., *Acta Chem. Scand.*, **8**, 266, 1059 (1954); **9**, 49 (1955).

<sup>10</sup> Irving, H., and Williams, R. J. P., *Nature*, **162**, 746 (1948).

<sup>11</sup> Irving, H., and Williams, R. J. P., *J. Chem. Soc.*, 3192 (1953).

### Determination of Strychnine in Nux Vomica by Paper Chromatography

Most methods used for the estimation of strychnine in nux vomica lead to gross inaccuracies due to brucine interference. Even oxidation of brucine with nitric acid under optimal condition (5 min. at 60° C.) leads to errors due to partial destruction of strychnine<sup>1</sup>. Gravimetric estimations also require amounts greater than 0.1 gm. in the sample<sup>1</sup>.

Paper chromatography has also been applied, but most of the methods have difficulty in separating brucine from strychnine. Dušinský and Tyllová<sup>2</sup> have overcome this difficulty by using nitric acid to oxidize brucine to o-brucichinone. This method probably suffers from errors outlined by de la Vega and Del Pozo<sup>1</sup>.

These can be obviated by using a simplified method. The aqueous sample was made alkaline with sodium hydroxide and the alkaloids were extracted with chloroform. The chloroform was then evaporated in a water-bath. The resulting alkaloids were dissolved in a measured volume of chloroform and aliquots of this extract, containing 60-20  $\gamma$  strychnine, were spotted on to Whatman No. 1 chromatography paper and developed overnight with *n*-butanol/*n*-propanol/0.05 N hydrochloric acid (1 : 2 : 1 v/v) at 20° C. by the ascending technique. The paper was dried and photographed in the ultra-violet to reveal the spots. The paper was then cut and eluted with water. The strychnine hydrochloride was estimated using a Beckman spectrophotometer model DU at  $\lambda_{\text{max}}$ . 255 m $\mu$ .<sup>3</sup>

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<sup>1</sup> de la Vega, F. A., and Del Pozo, A., *Galenica Acta (Madrid)*, **2**, 35 (1949), cited in *Chem. Abstr.*, **47**, 9571b (1953).

<sup>2</sup> Dušinský, G., and Tyllová, M., *Nature*, **181**, 1335 (1958).

<sup>3</sup> Kay, S., and Hoff, E. C., *J. Criminal Law, Criminol. Police Sci.*, **43**, 264 (1953), cited in *Chem. Abstr.*, **48**, 6325e (1954).

### Isolation of Piperonylic Acid from *Ocotea pretiosa*

As part of work on the chemical constituents of Brazilian Lauraceae<sup>1</sup>, we examined the benzene extracts (2.4 per cent) of the wood of *Ocotea pretiosa* (Nees) Mez, the sassafras tree of the State of Santa Catarina producing essential oil (chiefly safrole). Through extraction with aqueous bicarbonate solution a solid (comprising 0.03 per cent of the wood) was isolated and purified by crystallization from chloroform and vacuum sublimation. White crystals were obtained, melting point 227-29° C. (Kofler block). Determination of the melting point of a mixture and infra-red spectral comparison with an authentic sample established its identity with piperonylic acid.

Piperonylic acid is, apparently, quite rare in Nature. Its known occurrence is restricted to the plant family Lauraceae, where traces of it have been found accompanying relatively much larger amounts of substances containing the piperonyl moiety. Only once before<sup>2</sup> has it been isolated by solvent extraction from a natural source, the coto barks, where it occurs along with 6-piperonyl- $\alpha$ -pyrone (*Aniba coto* Kostermans) and accompanying 6-piperonyl- $\alpha$ -pyrone, 2,4-dimethoxy-6-hydroxy-3',4'-methyleneedioxybenzo-