

## Short Communications

### Normalized Projection Maps Calculated and Plotted by Computer

DIEGO FERRI,<sup>a</sup> OLOF WAHLBERG<sup>a,b</sup>  
and ERIK HÖGFELDT<sup>a</sup>

<sup>a</sup>Department of Inorganic Chemistry, The Royal Institute of Technology (KTH), S-100 44 Stockholm 70 and <sup>b</sup>Departments of Inorganic and Structural Chemistry, The Arrhenius Laboratory, P.O.B., S-104 05 Stockholm 50, Sweden

The theory of normalized projection maps was outlined in the 1950's by Sillén,<sup>1-3,5</sup> Biedermann,<sup>3</sup> and the Rossottis'.<sup>4-6</sup> The information about the average composition of complexes obtained by the MESAK method<sup>7</sup> combined with that from normal-

ized projection maps often gives the main species present in a given system. Further calculations with LETAGROP<sup>8-10</sup> gives an improved set of equilibrium constants and makes it possible to test the uniqueness of the model as well as the possible presence of minor constituents and systematic errors. An instructive example of what can be achieved by combining the MESAK method with projection maps can be found in a paper by Ingri.<sup>11</sup>

In order to simplify the use of normalized projection maps Ferri and Wahlberg<sup>12</sup> prepared an Algol program NORMEX for calculating such maps. The maps can be plotted using a line printer.

Photographic plots can also be obtained by using a program recently designed by Warnqvist.<sup>13</sup> NORMEX is constructed in a way that allows a high degree of freedom in the choice of normalizing conditions. At present the program is written for one, two, or three components, but an extension to four or more can easily be

\* Present address: Università di Napoli, Istituto Chimico, 80134 Napoli, Italy.

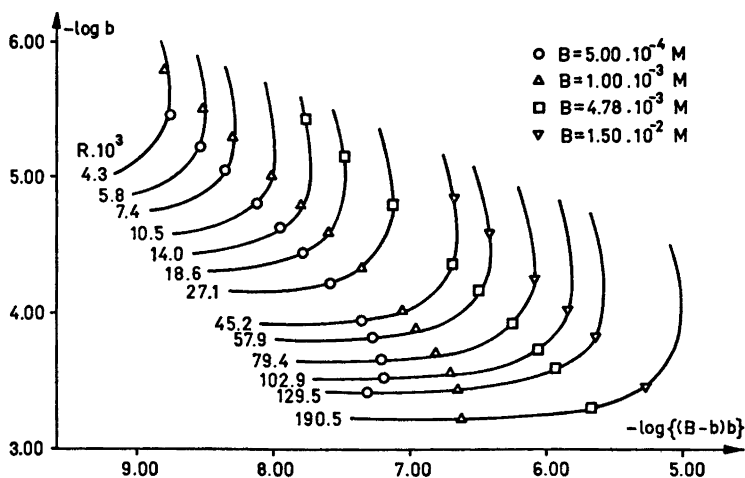


Fig. 1.  $-\log b$  plotted against  $-\log (B-b)b$  for the system In(III)-Cl<sup>-</sup> studies by Ferri.<sup>14</sup> The points are experimental and the curves computed with NORMEX and  $\log \beta_1 = 2.57$ ,  $\log \beta_2 = 3.84$ , and  $\log \beta_3 = 4.2$ .

made if the need arises. The use of NORMEX is illustrated in Fig. 1, where  $-\log b$  is plotted against  $-\log (B-b)b$  for the system  $\text{In}^{3+}-\text{Cl}^-$  in 3 M  $\text{NaClO}_4$  studied by one of us.<sup>14</sup>  $B$  is the total concentration of indium and  $b$  the concentration of free  $\text{In}^{3+}$  as measured by an indium amalgam electrode. The points are experimental and the curves have been computed with NORMEX using a model including the formation of  $\text{InCl}_2^+$ ,  $\text{InCl}_2^+$ , and  $\text{InCl}_3$  with  $\log \beta_1 = 2.57 \pm 0.02$ ,  $\log \beta_2 = 3.84 \pm 0.02$ , and  $\log \beta_3 = 4.2 \pm 0.1$ , which compares well with the values obtained by LETAGROP:  
 $\log \beta_1 = 2.58 \pm 0.01$ ,  $\log \beta_2 = 3.84 \pm 0.01$ ,  
 $\log \beta_3 = 4.25 \pm 0.02$

A detailed description of NORMEX and examples of how to use it are given in the paper by Ferri and Wahlberg,<sup>12</sup> and anyone interested can write to Dr. Wahlberg at the University of Stockholm for reprints and further information.

*Acknowledgements.* This work is part of a program supported by the Swedish Natural Science Research Council. Docent Derek Lewis revised the English text.

1. Sillén, L. G. *Acta Chem. Scand.* **10** (1956) 186.
2. Sillén, L. G. *Acta Chem. Scand.* **10** (1956) 803.
3. Biedermann, G. and Sillén, L. G. *Acta Chem. Scand.* **10** (1956) 203.
4. Rossotti, F. J. C. and Rossotti, H. S. *Acta Chem. Scand.* **9** (1955) 1166.
5. Rossotti, F. J. C., Rossotti, H. S. and Sillén, L. G. *Acta Chem. Scand.* **10** (1956) 203.
6. Rossotti, F. J. C. and Rossotti, H. S. *The Determination of Stability Constants*, McGraw, New York 1961.
7. Sillén, L. G. *Acta Chem. Scand.* **15** (1961) 1981.
8. Ingri, N. and Sillén, L. G. *Arkiv Kemi* **23** (1965) 97.
9. Sillén, L. G. and Warnqvist, B. *Arkiv Kemi* **31** (1969) 341.
10. Brauner, P., Sillén, L. G. and Whiteker, R. *Arkiv Kemi* **31** (1969) 377.
11. Ingri, N. *Acta Chem. Scand.* **17** (1963) 597.
12. Ferri, D. and Wahlberg, O. *Chem. Commun.*, Univ. Stockholm 1973, No. XII.
13. Warnqvist, B. *Personal communication* 1971.
14. Ferri, D. *Acta Chem. Scand.* **26** (1972) 733.

Received November 2, 1973.

## Polymannuronic Acid 5-Epimerase from the Marine Alga *Pelvetia canaliculata* (L.) Dcne. et Thur.

JOHN MADGWICK, ARNE HAUG and  
BJØRN LARSEN

*Institutt for marin biokjemi, N-7034  
Trondheim-NTH, Norway*

Polymannuronic acid 5-epimerase was first demonstrated as an extracellular product of the bacterium *Azotobacter vinelandii* (Lipmann).<sup>1</sup> Subsequent work dealt with characterisation of the enzymic epimerisation,<sup>2</sup> the nature of the alginate from *A. vinelandii*,<sup>3</sup> and the mechanism of the enzymatic reaction.<sup>4,5</sup> However, although it seemed likely that brown algae must contain a similar epimerase, it was hard to prove because these plants' enzymes are so difficult to extract due to the presence of large amounts of sulphated polysaccharides and phenolic compounds.<sup>6</sup> This communication presents evidence for an algal alginate epimerase and describes the preparation of a soluble enzyme system from the brown alga *P. canaliculata*. An ammonium sulphate fraction was obtained which catalysed the conversion of polymannuronic acid to a mixed polymer containing guluronic acid residues. The epimerisation reaction was revealed by changes in carbazole reactives<sup>7</sup> and with incorporation of tritium into the polyuronide fraction.<sup>4</sup>

*Materials and methods:* *P. canaliculata* was harvested from Lade and Flakk, Trondheimsfjord, on 18.5.1973 and 19.8.1973, respectively, and deep-frozen at  $-15^\circ\text{C}$ . A brei of *P. canaliculata* was made in water at  $0^\circ\text{C}$ , (800 ml/100 g wet wt.), using an Ultra-Turrax® blender. The material remaining after filtration with gauze (ca. 0.5 mm, 4 layers) was discarded. Centrifugation, (7000 g, 10 min,  $4^\circ\text{C}$ ), gave a pellet which was resuspended in iced water and resedimented. An acetone powder, (500 mg/100 g wet algae), was prepared from the washed pellet,<sup>8</sup> and subsequently extracted, (4 h,  $20^\circ\text{C}$ ), with a solution containing phosphate buffer, (pH 7.8, 0.05 M), NaCl, (0.2 M), and dithiothreitol ( $10^{-4}$  M). Solubles were separated by centrifugation, treated with ammonium sulphate at  $0^\circ\text{C}$ , and the precipitates between 40 % and 90 % saturation combined and dialysed against tris(hydroxymethyl)ami-