

precipitation from amorphous tall oil rosin (also a product of the above-mentioned company) and from a crystalline, continuously distilled product of Oulu Oy, Oulu). Bands typical of abietic acid occur at 8.66, 11.21, 12.65 and 13.87  $\mu$  and bands typical of dehydroabietic acid at 6.68, 8.82, 9.65, 12.20 and 13.89  $\mu$ . No bands are observed which would indicate the presence of tetrahydroabietic acid (characteristic bands at 8.47, 8.61 and 14.27  $\mu$ ), dihydroabietic acid (bands at 11.53 + 11.68 and 14.12  $\mu$ ; the shoulder at 14.12  $\mu$  results from the combined medium strong absorptions of abietic and dehydroabietic acids), dextropimaric acid (bands at 6.97, 11.75, 14.00 and 14.61  $\mu$ ), and *isodextropimaric acid* (bands at 8.00, 8.68 + 8.75, 11.00, 13.67 and 13.99  $\mu$ ). The rosin acid mixture isolated from tall oil rosin thus seems to consist solely of abietic and dehydroabietic acids. No signs indicating the presence of other acids of the abietic series or of pimaric acids are evident (if these do exist in the mixture, their contents amount to a few per cent at the most).

Absorption spectra \* recorded for binary mixtures of abietic and dehydroabietic acid suggested that the batch-distilled rosin acid samples studied contained about 55 % abietic acid and about 45 % dehydroabietic acid.

Absorption spectra \* recorded for rosin acid mixtures isolated by *cyclohexylamine* precipitation from crystalline tall oil rosin samples which were heated at 300°C for periods of different length revealed that the content of dehydroabietic acid in the mixtures increased with the time of heating. The rosin acid mixture precipitated with *cyclohexylamine* from a tall oil rosin sample that had been heated three hours at 300°C was found to consist almost solely of dehydroabietic acid. A similar observation concerning American tall oil rosin has been previously reported by Hasselström<sup>10</sup>.

The observation that the rosin acid mixture isolated from tall oil rosin does not contain pimaric acids is somewhat surprising and does not conform with the analytical data of Sandermann or with the general opinion of the composition of the rosin acid fraction of tall oil rosin. However, the somewhat erroneous conclusions that were drawn on the basis of the very extensive

work that was done after the turn of the century to determine the structure of pinabietic acid is now easily understood when one realizes that this rosin acid mixture primarily consisted of only two abietic acids.

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1. Juvonen, V.-V. *Finnish Paper Timber J.* **28** (1946) 8.
2. Johanson, S. *Svensk Papperstidn.* **53** (1950) 258.
3. Sandermann, W. *Fette und Seife* **49** (1942) 578.
4. Fleck, E. E. and Palkin, S. *J. Am. Chem. Soc.* **60** (1938) 921.
5. Mo och Domsjö Ab, Research Laboratory; cf. Johanson<sup>2</sup>.
6. Harva, O. *Paper and Timber, Finland* **34** (1952) 31.
7. Bruun, H. H. *Paper and Timber, Finland* **38** (1956) 577.
8. Bruun, H. H. *Paper and Timber, Finland* **39** (1957) No. 4.
9. Harris, G. C. and Sanderson, T. *J. Am. Chem. Soc.* **70** (1948) 334.
10. Hasselström, T. *Paper Trade J.* **118** (1944), TAPPI Sect., p. 134.

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## Association of Crystallized Bovine Plasma Albumin

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Many investigators have found that the molecular weight of bovine plasma albumin (B.P.A.) is about  $70 \times 10^5$ . This figure has been obtained from osmotic measurements (see, e. g., Scatchard

\* To be reproduced in the same series of papers as Refs. 7, 8

*et al.*<sup>1</sup>, and Adair and Robinson<sup>2</sup>). We have tried to reproduce these results by means of the osmometer described previously by two of the present authors<sup>3</sup>. By means of this apparatus it is possible to measure the osmotic pressure of the solution about one hour after it has been placed in the osmotic cell.

The preparation used was Armour crystallized B.P.A. The substance was dissolved in acetate buffer ( $M = 0.2$ ;  $\text{pH} = 4.64$ , the isoelectric point of B.P.A.) and put into the osmometer immediately after dissolution. Under these conditions we observed that solutions of B.P.A. showed an osmotic pressure corresponding to a molecular weight of  $270 \times 10^3$ . This result may be interpreted as an aggregation of four molecules of B.P.A.

In order to explain the disagreement between our results and those obtained by other workers it should be noticed that the experiments described by other investigators have been performed on stock solutions or by means of static-elevation cells, which require several days for the attainment of equilibrium. We suspected therefore that the disagreement was due to a gradual depolymerization of a tetramer. To verify this assumption the following experiments were performed. Two stock solutions of B.P.A. were prepared and placed at  $20^\circ\text{C}$  and  $30^\circ\text{C}$ , respectively. At certain intervals of time aliquots were withdrawn and their osmotic pressures determined at  $20^\circ\text{C}$ . The aliquot at  $30^\circ\text{C}$

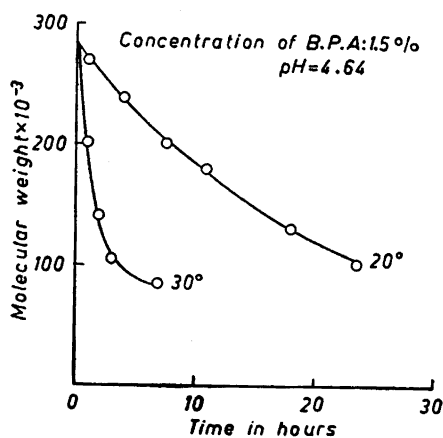


Fig. 1. Decrease in molecular weight with time at  $20^\circ\text{C}$  and  $30^\circ\text{C}$ .

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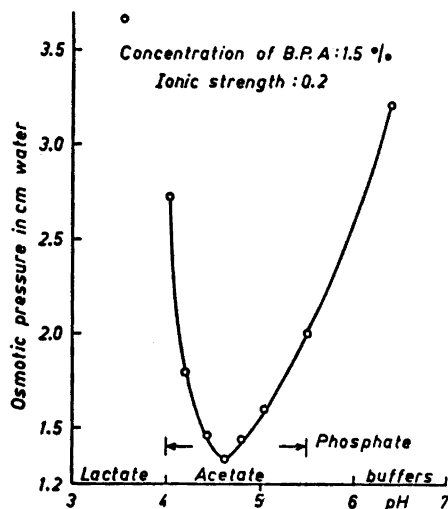


Fig. 2. Relation between osmotic pressure and pH.

was first cooled to  $0^\circ\text{C}$  to delay the reaction before the measurement. The results are in Fig. 1. It is seen that the molecular weight decreases with time, and that the rate of the decrease increases with temperature. These facts strongly indicate a gradual depolymerization.

Experiments have been carried out which show that the osmotic pressure, measured one hour after dissolution, is dependent on the pH. A minimum is found at the isoelectric point (Fig. 2).

It was further observed that the acidity of unbuffered solutions of B.P.A. increased with time. This phenomenon has been found previously by Tanford<sup>4</sup>.

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1. Scatchard, G., Batchelder, A. C. and Brown, A. *J. Am. Chem. Soc.* **68** (1946) 2320.
2. Adair, G. S. and Robinson, H. E. *Biochem. J. London* **24** (1930) 1864.
3. Christiansen, J. A. and Jensen, C. E. *Acta Chem. Scand.* **7** (1953) 1247.
4. Tanford, C. *J. Am. Chem. Soc.* **72** (1950) 441.

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