

lecithin and of crude brain there are considerable amounts of material (by weight) in this region. Our lecithin fractions have not been examined for choline, and the chromatograms of crude lipid in system *A* (Fig. 5) do not reveal choline in the very polar region, though from the behaviour during desalting it was concluded that a strong base was present. Choline might have been diluted out by other materials, present in larger amounts, in our crude brain lipid. Nevertheless, the sum of evidence suggests that Lovern's material differs substantially from ours, or alternatively that the lipids behave differently in the two solvent systems.

Regarding ethanolamine and serine phosphatides, Lovern's distributions (Lovern, 1952; Figs. 1 and 6) show, like our own (Fig. 5), that these two kephalins behave very similarly, with much overlapping of lipid fractions.

It was thought at the outset of this work that among the phosphatides the bases would be the main factor determining the distribution between solvent phases. This may be true when phosphatidylcholine is contrasted with phosphatidyl serine and ethanolamine. Among the kephalins, however, other factors such as the degree of unsaturation of the fatty acids, and length of the aliphatic chain may

be equally important. Moreover, we have some evidence that complex formation may occur between one lipid and another when their concentrations are high. In this circumstance two lipids may tend to move together and only separate at greater dilutions.

#### SUMMARY

1. An emulsion-resistant solvent system of water-methanol-carbon tetrachloride has been devised for the counter-current fractionation of brain lipids.

2. The behaviour of brain kephalin, lecithin, and mixed sphingomyelin and cerebroside has been described.

3. Modifications of the basic solvent system, for the fractionation of more polar lipids and for the separation of cholesterol and neutral fats from other lipids, are presented.

4. The relative positions of various known compounds in the counter-current distribution of crude brain lipid have been indicated.

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## A Nomogram for Ammonium Sulphate Solutions

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In the purification of enzymes and other proteins it is frequently necessary to fractionate with ammonium sulphate by collecting the fraction which is precipitated on passing from one given percentage saturation to another. One is then faced with the problem of calculating how much solid ammonium

sulphate must be added to a given volume of a solution of a known degree of saturation to bring it to a certain desired degree of saturation.

Such a calculation from the physical tables is distinctly laborious, owing to the form in which the data are given. The main difficulty is caused by the

fact that, owing to change of volume, the degree of saturation is far from being proportional to the amount of the salt added to a given volume of water.

lines. This shows that 500 ml. of a 0.25-saturated solution requires the addition of 193 g. of solid ammonium sulphate to bring it to 0.8-saturation. It must be noted that the final scale reads downwards.

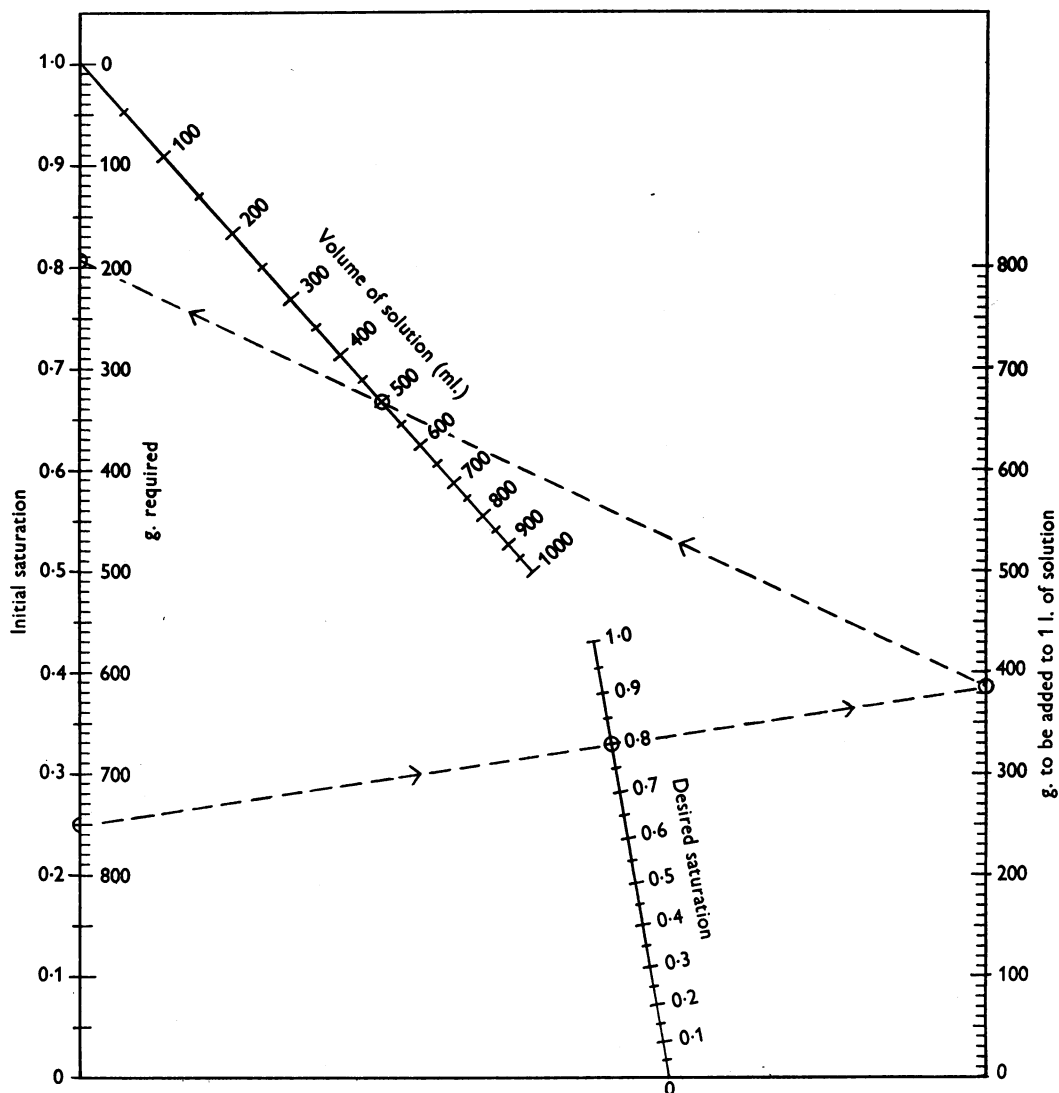


Fig. 1. A straight line through the initial saturation and the desired saturation gives the amount of solid  $(\text{NH}_4)_2\text{SO}_4$  to be added to 1 l. of the solution. A line from this point passing through the volume of the solution gives the amount required.

The nomogram shown in Fig. 1, which is self-explanatory, avoids the necessity for calculation. An example of its use is indicated by the dotted

Full saturation is taken as being given by adding 760 g. to 1 l. of water, i.e. as saturation at room temperature.