

mary phases are  $(\text{Na}_2, \text{NaLi})\text{SiO}_3$  solid solutions,  $\text{Li}_2\text{SiO}_3$ ,  $\text{Na}_2\text{Si}_2\text{O}_5$  and  $\text{Li}_2\text{Si}_2\text{O}_5$ , all three of which form solid solutions of limited extent, and the three modifications of silicon dioxide, namely, quartz, tridymite, and cristobalite. The liquidus fields meet at two ternary eutectics: one at  $697^\circ$ , with  $(\text{Na}_2, \text{NaLi})\text{SiO}_3$ ,  $\text{Li}_2\text{SiO}_3$  and  $\text{Na}_2\text{Si}_2\text{O}_5$ , the other at  $637^\circ$ , with  $\text{Li}_2\text{Si}_2\text{O}_5$ ,  $\text{Na}_2\text{Si}_2\text{O}_5$  and quartz as the eutectic constituents.  $\text{Li}_2\text{Si}_2\text{O}_5$  melts incongruently throughout its region of existence in the system, the reaction temperature descending from  $1033^\circ$  in the binary system,  $\text{Li}_2\text{SiO}_3$ - $\text{SiO}_2$ , to  $641^\circ$ , the peritectic end-point in the ternary system, with  $\text{Li}_2\text{Si}_2\text{O}_5$ ,  $\text{Li}_2\text{SiO}_3$ ,  $\text{Na}_2\text{Si}_2\text{O}_5$  and liquid in coexistence.

The inversion temperature of quartz and tridymite has been redetermined. The temperature of  $870 \pm 10^\circ$ , given by Fenner in 1913, is confirmed, the value obtained being  $867 \pm 3^\circ$ .

Refractive indices of glasses of various compositions in the system were measured.

A discussion of solid solution relationships of sodium and lithium compounds in general is given, with particular reference to the theoretical aspects of the subject.

Minor revisions of the phase relations in the systems,  $\text{Na}_2\text{SiO}_3$ - $\text{SiO}_2$  and  $\text{Li}_2\text{SiO}_3$ - $\text{SiO}_2$ , particularly with respect to the polymorphic behavior of  $\text{Na}_2\text{Si}_2\text{O}_5$  and  $\text{Li}_2\text{Si}_2\text{O}_5$ , are presented.

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### Pressure-Volume-Temperature Relations in Solutions. III. Some Thermodynamic Properties of Mixtures of Aniline and Nitrobenzene

BY R. E. GIBSON AND O. H. LOEFFLER

A few months ago we noted<sup>1</sup> that the absorption of light by mixtures of aniline and nitrobenzene was strongly influenced by pressure changes. In investigating this effect we thought it desirable to examine thoroughly the volume changes which take place when these two liquids are mixed in different proportions at different pressures and temperatures. Furthermore, we have noticed in the pure liquids<sup>2</sup> that, whereas the internal pressure  $(\partial E/\partial V)_T$  varies with temperature at constant volume, another quantity,  $P_A$ , computed from the  $P$ - $V$ - $T$  data is independent of temperature at constant volume and is expressible as  $a'/V^n$ . We have tentatively identified  $P_A$  as the attractive internal pressure of the liquid. In this paper we shall give data from which the volumes of aniline-nitrobenzene mixtures may be determined at all concentrations, between  $25$  and  $85^\circ$  and between  $1$  and  $1000$  bars, and we shall examine the behavior of  $P_A$  in these solutions.

#### Experimental Results

The solutions were all made up independently from weighed amounts of samples of aniline and nitrobenzene purified to a degree which already has been described.<sup>2</sup> The specific volumes of

these solutions were determined at  $25.00^\circ$  in 55-ml. U-tube pycnometers, and the thermal expansions were measured<sup>3</sup> at  $10^\circ$  intervals from  $25$  to  $85^\circ$  in a weight dilatometer of 18 ml. capacity made of vitreous silica.

The volumes of the solutions were expressed as functions of the temperature by empirical equations of the form

$$v = v_{55} + a(t - 55) + b(t - 55)^2 + c(t - 55)^3 \quad (1)$$

We already have published for the pure components such equations whose coefficients we may call  $a_1^0$ ,  $a_2^0$ ,  $b_1^0$ ,  $b_2^0$ , etc. From them we computed<sup>4</sup>  $\Delta_T v_1^0$  and  $\Delta_T v_2^0$  at each temperature, using  $55^\circ$  as the base temperature, and thence the quantity  $\Delta_T v - (x_1 \Delta_T v_1^0 + x_2 \Delta_T v_2^0) \equiv \delta_T$ . This function measures the departures of the thermal expansions

(3) For details of the procedure in these measurements the reader is referred to the second paper of this series (ref. 2).

(4) The symbols used here are as follows. The subscripts 1 and 2 refer to nitrobenzene and aniline in solution, respectively, the superscript 0 implies the pure components, and symbols without subscripts refer to the solutions. The weight and mole fractions are given by  $x$  and  $X$ , respectively,  $R_2 = X_2/X_1$ , and  $v$  and  $V$  mean the specific and molal volumes. For a solution  $V = v/(x_1/M_1 + x_2/M_2)$ ,  $M$  being the molecular weight.  $\Phi$  is the apparent molal volume of a component.  $\Delta v$  and  $\Delta V$  are the volume changes on mixing per gram and per mole of solution. All volumes are given in milliliters.

The pressure in kilobars is given by  $P$ , the temperature by  $t$  (centigrade scale) or  $T$  (absolute), and the total energy by  $E$ .  $\Delta T$  and  $\Delta P$  denote the finite changes with temperature and pressure, respectively, of the quantities to which they are prefixed.  $h = -\Delta P_0/v_P = 0$ .  $z_1 = 1/(1 + R_2 V_2^0/V^0)$ ,  $z_2 = (1 - z_1)$  are the volume fractions of the components.

(1) R. E. Gibson and O. H. Loeffler, *J. Phys. Chem.*, **43**, 207 (1939).

(2) R. E. Gibson and O. H. Loeffler, *THIS JOURNAL*, **61**, 2515 (1939).

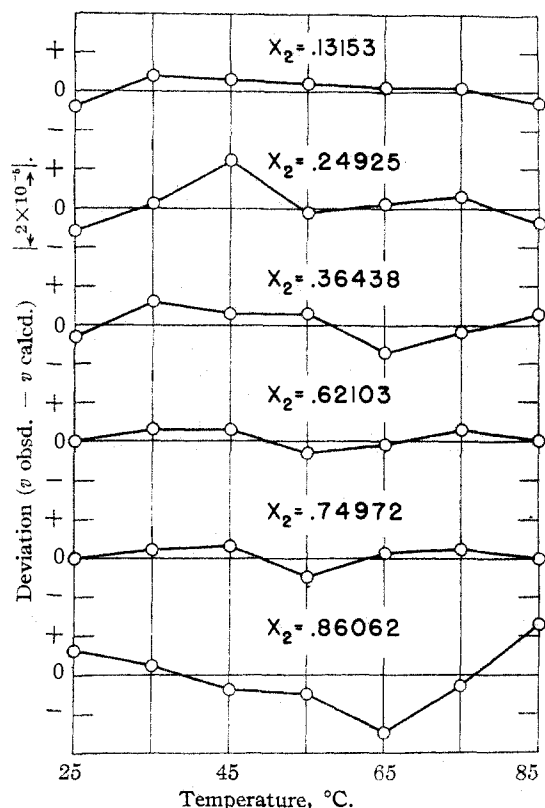


Fig. 1.—Deviations of the values of the specific volumes of aniline-nitrobenzene solutions as calculated from equation (1) from the observed ones at different temperatures.

from the ideal law of mixtures, and it may be represented accurately by the relation

$$\delta v = l + m(t - 55) \quad (2)$$

In these solutions  $(x_1 \Delta_T v_1^0 + x_2 \Delta_T v_2^0)$  is always numerically greater than  $\Delta_T v$ . The coefficients  $l$  and  $m$  were determined by a simplified method of least squares.<sup>5</sup> It will be seen that the coefficients in equation (1) were therefore obtainable from the following relations

$$\begin{aligned} a &= x_1 a_1^0 + x_2 a_2^0 + m; & b &= x_1 b_1^0 + x_2 b_2^0 \\ c &= x_1 c_1^0 + x_2 c_2^0 & v_{\text{obsd.}} &= v_{\text{obsd.}} + l \end{aligned}$$

The coefficients  $a$ ,  $b$ ,  $c$ , and  $m$  are given in Table I and the deviation curves in Fig. 1 indicate how well the equations fit the data. It is of interest to note that the coefficients of the second and third powers of the temperature are found directly from the simple law of mixtures.

The compressions of the solutions from 1 to 500 and from 1 to 1000 bars were measured in exactly the same way as those of the pure com-

(5) See Ostwald-Luther, "Physikochemischer Messungen," 4th ed., Akademische Verlagsgesellschaft, Leipzig, 1925, p. 39.

TABLE I

COEFFICIENTS IN THE EQUATIONS REPRESENTING THE SPECIFIC VOLUMES OF ANILINE-NITROBENZENE MIXTURES AS FUNCTIONS OF THE TEMPERATURE (EQUATION 1)

The last column gives the value of the principal coefficient in equation (2).

Weight fraction of aniline $x_2$	Mole fraction of aniline $X_2$	$v_{55}$	$10^3 a$	$10^3 b$	$10^3 c$	$10^3 m$
0.0000	0.0000	0.85575 <sub>1</sub>	7.274	6.931	1.528	
.13153	.16684	.87716 <sub>0</sub>	7.465	7.241	1.601	-0.0193
.24925	.30306	.89597 <sub>4</sub>	7.643	7.519	1.666	-0.0303
.36438	.43117	.91410 <sub>4</sub>	7.819	7.791	1.730	-0.0382
.49517 <sup>a</sup>	.56461	.93431 <sub>1</sub>	8.025	8.100	1.802	-0.0413
.62103	.68422	.95339 <sub>9</sub> <sup>b</sup>	8.218	8.398	1.873	-0.0501
.74972	.79842	.97265 <sub>2</sub>	8.425	8.701	1.944	-0.0483
.86062	.89088	.98885 <sub>0</sub>	8.615	8.963	2.006	-0.0358
1.0000	1.0000	1.00874 <sub>6</sub>	8.874	9.292	2.083	

<sup>a</sup> The thermal expansions of this solution were measured only to 65°, and the compressions only at 25°. <sup>b</sup> Mean of two entirely independent determinations.

ponents<sup>2</sup> and the relative volume changes were expressed as functions of the pressure by the Tait equation

$$k = C \log [(B + P)/(B + P_0)] \quad (3)$$

where  $C$  has the same value as for aniline and nitrobenzene, *viz.*, 0.21591 at all temperatures. The constant  $B$  was determined at each temperature from the compressions to 1000 bars. The compressions to 500 bars computed by the appropriate Tait equation always agreed with the observed ones within experimental error. The compressions were measured at 25, 45, 65, and 85°, and the constant  $B$  was expressed as a function of the temperature by equations of the type

$$B = B_{25} + \alpha(t - 25) + \beta(t - 25)^2 \quad (4)$$

which were fitted to the data at 25, 45, and 85°. In Table II we give the values of  $B_{25}$ ,  $\alpha$  and  $\beta$  for

TABLE II

COEFFICIENTS IN EQUATION (4) REPRESENTING THE CONSTANT  $B$  IN THE TAIT EQUATION AS A FUNCTION OF TEMPERATURE

$x_2$	$B_{25}$ (kilobars)	$10^3 \alpha$	$10^3 \beta$	$10^3 B$ (obsd. - calcd.) at 65°
0.0000	1.8652	9.62	1.48	+0.2
.13153	1.8740	9.83	1.72	- .2
.24925	1.8843	9.96	1.86	+2.7
.36438	1.8975	10.08	1.97	+1.2
.62103	1.9320	10.09	1.64	-1.1
.74972	1.9536	10.21	1.62	-3.6
.86062	1.9768	10.48	1.82	+1.6
1.0000	2.0066	10.81	1.99	-0.3

the various solutions together with the differences between the observed and calculated values of  $B$  at 65° as an illustration of the fit of the empirical equations.

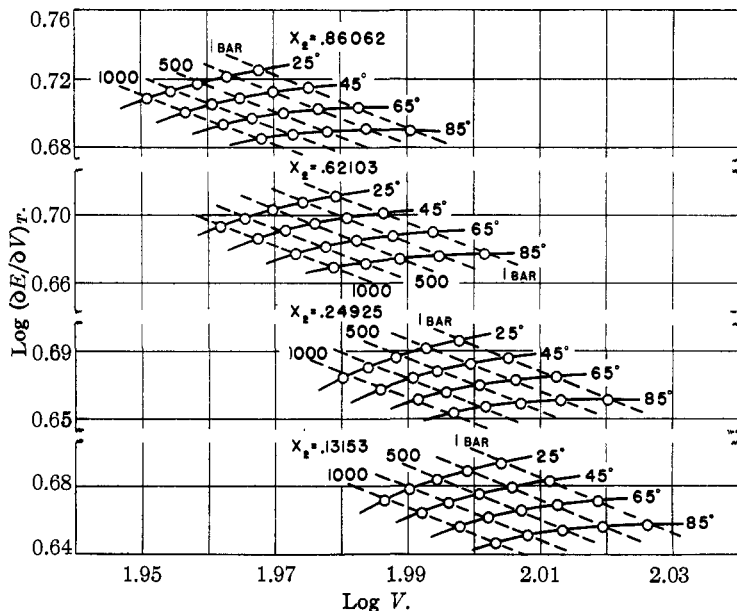


Fig. 2.—The logarithm of the internal pressure  $(\partial E/\partial V)_T$  of some aniline-nitrobenzene solutions as a function of the logarithm of the molal volume. Note the different curves for the different temperatures.

**The Internal Pressures  $(\partial E/\partial V)_T$  and Related Quantities**

From equations (1) and (3) the expansion coefficients  $(\partial v/\partial T)_P$  and the compressibilities were computed at different pressures and temperatures<sup>6</sup> and hence the coefficients  $(\partial P/\partial T)_V$  and  $(\partial E/\partial V)_T$  were calculated readily for each solution at 25, 45, 65, and 85°, and at 1, 250, 500, 750, and 1000 bars. The curves of  $(\partial E/\partial V)_T$  vs.  $V$ , or of  $(\partial P/\partial T)_V$  vs.  $V$ , for the solutions resembled those for the pure liquids in all major respects<sup>2</sup>; in particular,  $(\partial E/\partial V)_T$  decreased with rise of temperature at constant volume. A sample of our results illustrating this temperature effect is given in Fig. 2 where  $\log (\partial E/\partial V)_T$  is plotted against  $\log V$  for several solutions.

Applying the method of analysis that we developed for the pure liquids to the results for the solutions,<sup>2</sup> we computed the quantity  $(T(\partial P/\partial T)_V + B) = P_A$  which we have interpreted as the attractive internal pressure set up by the intermolecular forces,  $(B + P)$  being assumed to be a measure of the repulsive pressure. In Fig. 3 we have

plotted  $\log P_A$  vs.  $\log V$  for all the solutions including the end members, and in order to facilitate a comparison of Figs. 2 and 3, we have used the same scale in both diagrams. It will be seen that in Fig. 3 the points for the different temperatures all fall on the same straight line for each solution and that the lines for the different solutions all have the same slopes. Over the region we have studied we may, therefore, write

$$P_A = a'/V^{2.737} \quad (5)$$

2.737 being the constant slope of the lines in Fig. 3. The constants  $a'$  were determined from the data and average values are given in Table III. It should be noted that the points at 65 and 85° for nitrobenzene lie on a line which is slightly below that drawn through the points for the lower temperatures. We are not certain that

this deviation is a real one and that it does not arise in the equations used for computing the derivatives; but it is significant that the same deviations are found in the solutions rich in nitro-

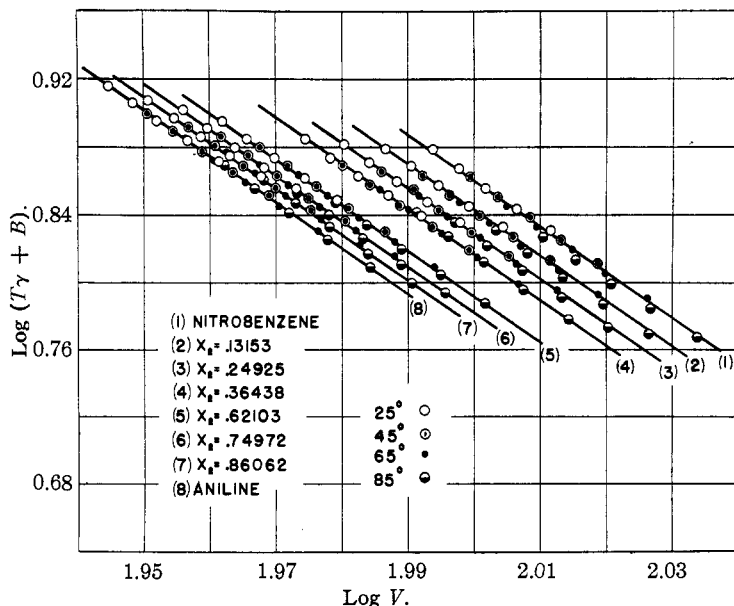


Fig. 3.—The logarithm of the attractive internal pressure  $P_A = (T\gamma + B)$  of aniline-nitrobenzene solutions as a function of the logarithm of the molal volume. Note that the points for different temperatures fall on the same line for each solution. The scale in this diagram is the same as in Fig. 2.  $\gamma$  represents the coefficient  $(\partial P/\partial T)_V$ .

benzene and gradually vanish as the concentration of aniline increases.

(6) For details of these calculations, see ref. 2.

TABLE III

THE GENERALIZED FRACTION  $y_1$ , AND A COMPARISON OF THE VALUES OF  $a'$  DETERMINED DIRECTLY FOR EACH SOLUTION (EQUATION 5) AND THE VALUES OF  $a'$  COMPUTED FROM EQUATION (9)

$x_2$	$y_1$	$10^6 a'$ (obsd.)	$10^6 a'$ (calcd.)	$\Delta a' \times 10^6$ at 25°
0.0000	1.0000	2.163		
.13153	0.90398	2.071	2.079	134
.24925	.81114	2.004	2.010	218
.36438	.71325	1.947	1.951	272
.62103	.46527	1.840	1.841	279
.74972	.32250	1.797	1.796	235
.86062	.18761	1.766	1.764	155
1.0000	.0000	1.730		

### The Variation of $P_A$ with Concentration

We shall now attempt to calculate  $P_A$  and  $a'$  for the solutions from a knowledge of the properties of the pure components. If  $V$  is the volume of 1 mole of solution,<sup>4</sup>  $V^0$  the volume of the corresponding amounts of the components (*i. e.*, the ideal molal volume of the solution) and if  $P_A$  is the observed attractive pressure in the solution, then  $P_A^0$ , the attractive pressure of the solution at volume  $V^0$ , is given by

$$P_A^0(V^0)^{2.737} = P_A V^{2.737} = a' \quad (6)$$

If, furthermore, we suppose the attractive pressures set up between like molecules in the solution to be the same as in the pure liquids, *viz.*,  $P_{11}$  for nitrobenzene and  $P_{22}$  for aniline, and call the attractive pressure between the unlike molecules  $P_{12}$ , we may write

$$P_A^0 = y_1^2 P_{11} + y_2^2 P_{22} + 2y_1 y_2 P_{12} \quad (7)$$

where  $y_1$  and  $y_2$  are concentration functions that indicate the probabilities of significant interactions between the molecules,<sup>7</sup> and are defined as  $y_1 = 1/(1 + R_2 U_2/U_1)$  and  $y_2 = (1 - y_1)$ . The parameter  $U_2/U_1$  may be regarded as an empirical constant which is equal to unity when  $y$  is the mole fraction and is  $V_2^0/V_1^0$  when  $y$  is the volume fraction.

When the mole fractions or the volume fractions were used in equation (7) no results of any special interest were found, and the computed values of  $P_{12}$  varied with concentration. We,

(7) This treatment, which partly follows that proposed by Scatchard [*Chem. Rev.*, **8**, 321 (1931)], does not limit  $y_1$  and  $y_2$  to the volume fractions based on the volumes of the liquids under ordinary conditions. We also wish to point out that our method of analysis implies that  $P_A$  depends chiefly on the mean distance between the molecules and is not greatly influenced by their distribution in the liquid, whereas the repulsive internal pressure depends strongly on the distribution of the molecules in the liquid. This seems plausible because the attractive forces between the molecules vary much less rapidly with distance than do the repulsive forces.

therefore, assumed<sup>8</sup> that  $P_{12}$  was equal to a constant, *viz.*,  $(P_{11}P_{22})^{1/2}$ , and, using the relation

$$P_A^0 = (y_1 P_{11}^{1/2} + y_2 P_{22}^{1/2})^2 \quad (8)$$

we computed  $U_2/U_1$  from our results at 25° and 1 bar and found it to be practically independent of concentration having the average value 0.5263.<sup>9</sup>

Combining equations (5), (6), and (8) we get equation (9)

$$a' = \left\{ y_1 a_{11}^{1/2} \left( \frac{V^0}{V_1^0} \right)^{1.368} + y_2 a_{22}^{1/2} \left( \frac{V^0}{V_2^0} \right)^{1.368} \right\}^2 \quad (9)$$

which should enable us to calculate  $a'$  for any solution in terms of the constants  $a_{11}$ ,  $a_{22}$ ,  $V_1^0$ , and  $V_2^0$  for the pure liquids, the *ideal* molal volume of the solution and the fraction  $y_1$  which contains the disposable constant  $U_2/U_1$ . In Table III we list values of  $y_1$  computed on the assumption that  $U_2/U_1 = 0.5304$  (the reason for the choice of this figure, which differs slightly from that given above, will appear under the discussion of the volume change on mixing) together with the values of  $a'$  obtained directly from our data for the solutions and values of  $a'$  computed from equation (9) when the volumes at 25° and 1 bar were used. The agreement between these two series of figures indicates that equation (9) does give an adequate representation of  $a'$  as a function of concentration. Moreover,  $V^0/V_1^0$  changes very little with pressure and temperature, the extreme variation over the range we have studied being less than 0.4%. We may, therefore, conclude that  $y_1$ ,  $y_2$  and hence  $U_2/U_1$  are independent of pressure and temperature within the limits of accuracy of our results.

### The Volume Changes on Mixing

The changes in volume which occur when the components are mixed in different proportions at different pressures and temperatures may be expressed conveniently in terms of the apparent

(8) See G. Scatchard, *Trans. Faraday Soc.*, **33**, 162 (1937); S. E. Wood, *THIS JOURNAL*, **59**, 1511 (1937); J. H. Hildebrand and J. M. Carter, *ibid.*, **54**, 3599 (1932).

(9) In terms of the principle of independent surface or group action [I. Langmuir, *Colloid Symposium Monograph*, **III**, 48 (1925); J. A. V. Butler, D. W. Thomson, and W. H. MacLennan, *J. Chem. Soc.*, 674 (1933)]  $y_1$  and  $y_2$  may be regarded as the volume fractions of the active groups in the nitrobenzene and aniline molecules. An examination of the parachors of benzene, nitrobenzene, and aniline suggests that the molal volume of the  $\text{NH}_2$  group is approximately one-half that of the  $\text{NO}_2$  group (Landolt-Börnstein, "Tabellen," *Ergänzungsband II(a)*, p. 178). Roughly the same result is obtained if one compares the molal volumes of benzene, nitrobenzene, and aniline under pressures where the liquids have the same compressibilities (ref. 1). There is, therefore, some justification for associating  $y_1$  and  $y_2$  with the volume fractions of the  $\text{NO}_2$  and  $\text{NH}_2$  groups in the solutions.

volumes of one of the components. It will be recalled that if

$$\Delta V = V - (X_1V_1^0 + X_2V_2^0) \text{ then } \Delta V = X_2(\Phi_2 - V_2^0)$$

In the aniline-nitrobenzene system an expansion occurs when the components are mixed,  $(\Phi_2 - V_2^0)$  being always positive. Increase of pressure and of temperature both diminish  $(\Phi_2 - V_2^0)$ , an observation to which special attention is drawn.

A clue to the origin of these volume changes may be obtained from an examination of the effect of temperature on the volume changes on mixing when the volumes of the unmixed components are kept constant. In Fig. 4 we have plotted  $\Delta V$  against  $V^0$  for various solutions. It will be seen that at constant volume, and presumably constant attractive pressure,  $\Delta V$  decreases quite rapidly as the temperature rises, and that this decrease is roughly proportional to  $\Delta V$  itself. Having in mind the interpretation we gave of similar changes in the  $(\partial E / \partial V)_T - V$  curves, we suggest the following explanation of this decrease of  $\Delta V$  with rise of temperature at constant  $V$ . We assume that the change with temperature in the randomness of the distribution of the molecules in a liquid is proportional to the order already existing in the liquid, and that in a solution of aniline in nitrobenzene the molecular distribution is more random than in the pure liquids at the same temperature (the increase in disorder determining to some extent the volume change on mixing). We should, therefore, expect that a given rise of temperature would have a greater disordering effect on the unmixed liquids than on the solution, and that at high temperatures the change in randomness on mixing should approach zero, and the volume change on mixing should decrease accordingly. The change of  $(B + P)$  with temperature at constant volume has been associated<sup>2</sup> with the change in the repulsive pressure due to increase in the randomness of the molecular distribution. Our data show that  $(B + P)_V$  increases more rapidly with temperature for the pure liquids than for the solutions.

In a recent paper<sup>1</sup> we attempted to express  $\Delta v$  in terms of the net internal (or repulsive) pres-

ures  $B'$  of the solutions and  $B_1$  and  $B_2$  of the pure components by the relation

$$\Delta v / C(x_1v_1^0 + x_2v_2^0) - (z_1 \log B_1 + z_2 \log B_2) = \log B' \quad (10)$$

It was assumed that, on forming the solution at atmospheric pressure, the pure components first expanded or contracted until their repulsive pressures came to the common value  $B'$  and then mixed without further change. We indicated also that the values of  $\log B'$  computed from the observed values of  $\Delta v$  by means of equation (10) were *smaller* than the observed values of  $\log B'$ . As equation (10) represents the volume change on mixing on the assumption that mixing introduces no change in the distribution of the molecules, it is suggested that the discrepancy

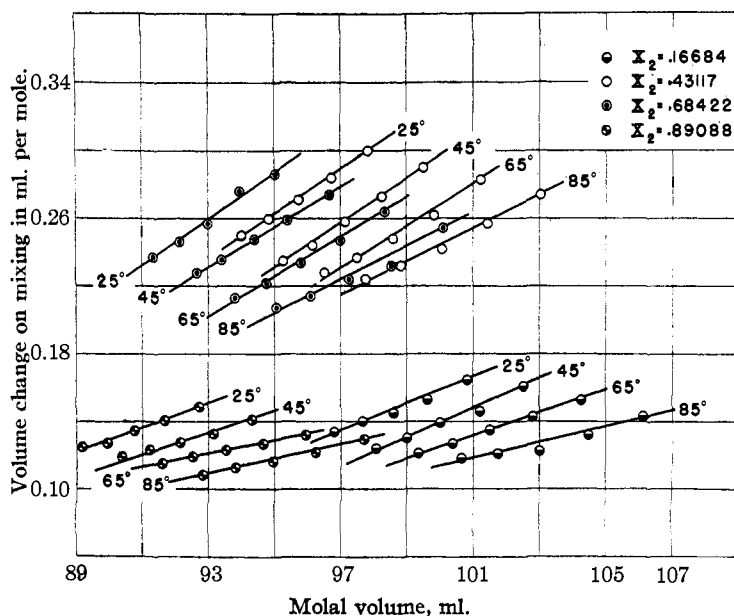


Fig. 4.—The volume changes on mixing per mole of solution in four solutions of aniline in nitrobenzene as functions of the volume at different temperatures.

between the observed and calculated values of  $B'$  is due to an increase in randomness on mixing. In other words, on the formation of the actual solution a given volume increase ( $\Delta v$ ) results in a smaller decrease in  $\log B'$  than would be expected if no increase in randomness accompanied the mixing of the components. It is of interest to note that the values of  $B'$  computed by equation (10) approach the observed ones more closely as the temperature is raised. These effects are small but they contribute qualitatively to the evidence correlating an increase in the randomness of the molecular distribution with an increase in the

quantity  $B'$  or  $(B + P)$  which we have tentatively identified as the repulsive internal pressure of the liquid or solution.

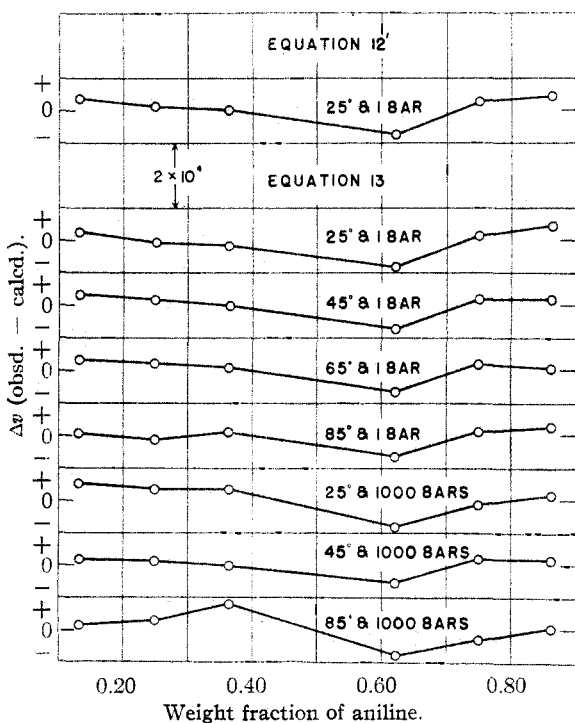


Fig. 5.—Deviations of the values of the volume change on mixing in aniline-nitrobenzene solutions as computed from equation (12') or (13) from the observed values at different pressures, temperatures and concentrations.

In order to represent the volume changes on mixing as functions of the composition of the solutions, we used equations of the type<sup>10</sup>

$$\Delta V/(X_1V_1^0 + X_2V_2^0) = pz_1z_2(1 + qz_2^3) \quad (11)$$

where  $z_1$  and  $z_2$  are the volume fractions, to represent our data. At 25° with  $p = 0.01230$  and  $q = 0.341$ , equation (11) reproduced the results quite well, the average deviation and the largest deviation corresponding to errors of 1.6 and  $3 \times 10^{-5}$ , respectively, in the specific volume. Equally satisfactory fits were obtained at the other temperatures and pressures. The term  $qz_2^3$  in equation (11) shows that the curve of  $\Delta V/(X_1V_1^0 + X_2V_2^0)$  vs.  $z_1$  is unsymmetrical, the maximum lying between  $z_1 = 0.5$  and  $z_1 = 0$ . As this asymmetry is found in the effect of concentration on the absorption of light and on the attractive internal pressures of mixtures of nitrobenzene and aniline, it is of interest to examine equations which express the dependence of  $\Delta V$  on concentration in

(10) G. Scatchard, S. E. Wood and J. M. Mochel, *J. Phys. Chem.*, **43**, 122 (1939).

terms of the general fractions  $y_1$  and  $y_2$ . It should also be remarked that the usefulness of equation (11) is diminished somewhat by the fact that  $z_1$ ,  $z_2$ ,  $p$ , and  $q$  all vary with temperature and pressure.

A trial of equation (12) (or its equivalent form equation 12') which is analogous to equation (11) showed that it gave a good but not altogether satisfactory representation of our data.<sup>11</sup>

$$\Delta V/(X_1U_1 + X_2U_2) = Ky_1y_2 \quad (12)$$

$$(\Phi_2 - V_2^0) = ky_1 = k/(1 + R_2U_2/U_1) \quad (12')$$

The constants  $k$  and  $U_2/U_1$  in equation (12') determined from the data by the method of least squares<sup>12</sup> are given in Table IV and the differ-

TABLE IV

THE CONSTANTS  $k$  AND  $U_2/U_1$  AS OBTAINED BY THE METHOD OF LEAST SQUARES FROM EQUATION (12') REPRESENTING THE VOLUME CHANGE ON MIXING AS A FUNCTION OF THE GENERALIZED VOLUME FRACTIONS AT DIFFERENT PRESSURES AND TEMPERATURES

$t$ , °C.	$P$ (bars)	$k$	$U_2/U_1$	Average $\Delta v$ (obsd. - calcd.) $\times 10^5$
25	1	1.0428	0.6600	3.3
45	1	1.0297	.6922	2.8
65	1	1.0077	.7164	2.2
85	1	0.9826	.7299	2.4
25	1000	.8694	.6625	3.0
45	1000	.8023	.6265	2.4
65	1000	.7854	.6535	3.8
85	1000	.7649	.6746	3.0

ences between the observed and calculated values of  $\Delta v$  at 25° and 1 bar are plotted in Fig. 5. The largest deviation corresponds to an error of  $-7 \times 10^{-5}$  in  $\Delta v$ . The deviation curves for the results at the other temperatures and pressures are the same as the curve we have given in Fig. 5 within an error of  $2 \times 10^{-5}$  in  $\Delta v$ . In order to give an idea of the numerical value of  $\Delta v$  we have recorded  $\Delta v$  at 25° and 1 bar in the last column of Table III. The parameter  $U_2/U_1$  as obtained from equation (12) varies greatly with temperature and is considerably larger than we found from the attractive pressure equation. In view of our discussion of the effect of change in molecular distribution on the volume change on mixing, of the complicated nature of this quantity, and of the fact that the compressibility of the solution is an important factor in determining the volume

(11) From equation (12) it may be seen that  $(V_2 - V_2^0) = ky_1^2$ ,  $V_2$  being the partial molal volume. The analogy with Hildebrand's equation for regular solutions is evident [THIS JOURNAL, **51**, 78 (1929)]. Furthermore, as  $y_1 = 1/(1 + R_2U_2/U_1)$ , this is the same type of equation that was proposed by van Laar [*Z. physik. Chem.*, **72**, 723 (1910)].

(12) We are indebted to Dr. W. Edwards Deming of the U. S. Department of Agriculture for valuable suggestions concerning these calculations.

change on mixing,<sup>13</sup> it seemed legitimate to regard equation (12) as too simple to be of much physical significance and to examine the behavior of  $U_2/U_1$  in some empirical equations.

The parameters  $k_1$  and  $U_2/U_1$  in equation (13) were evaluated by a least squares method at 25, 45, 65, and 85° both at 1 and 1000 bars.

$$(\Phi_2 - V_2^0) = V k_1 y_1 / \log (B' + P) \quad (13)$$

The results are given in the third and fourth columns of Table V. Although  $U_2/U_1$  ob-

TABLE V

THE CONSTANTS  $k_1$  AND  $U_2/U_1$  IN EQUATION (13) AS OBTAINED BY THE METHOD OF LEAST SQUARES AT DIFFERENT TEMPERATURES AND PRESSURES. IN THE LAST COLUMN ARE GIVEN AVERAGE VALUES OF  $k_1$  CONSISTENT WITH A VALUE OF  $U_2/U_1$  INDEPENDENT OF PRESSURE AND TEMPERATURE

$t$ , °C.	$P$ (bars)	$U_2/U_1$	$k_1 \times 10^3$	$\frac{k_1 \times 10^3}{(U_2/U_1 = 0.5304)}$
25	1	.5207	2.744	2.780
45	1	.5346	2.201	2.199
65	1	.5405	1.668	1.657
85	1	.5258	1.152	1.162
25	1000	.5609	4.033	3.940
45	1000	.5308	3.444	3.454
65	1000	.5530	3.095	3.055
85	1000	.5755	2.766	2.669

tained in this way is very sensitive to small fluctuations in the specific volume-concentration data, the values we obtained were practically constant over the whole temperature range at 1 bar, and varied no more at 1000 bars than might be expected from the greater uncertainty of the experimental data at the higher pressures. The average value of  $U_2/U_1$  at 1 bar, which was found to be 0.5304, was assumed to be a constant characteristic of the liquid mixture and independent of the pressure, temperature, and concentration. From it we computed the values of  $y_1$  given in Table III. In the fifth column of Table V we give the average values of  $k_1$  consistent with  $U_2/U_1 = 0.5304$ ; and in Fig. 5 we have plotted the differences between the observed values of  $\Delta v$  and those computed from equation (13) with these adjusted values of  $y_1$  and  $k_1$ . The deviation curves for the different pressures and temperatures being very much alike, we have given only a few examples. These curves indicate that equation (13) does not represent the volume change on mixing as well as might be desired; indeed, the deviations are only slightly smaller than those given by equation (12'). There is always a de-

viation of approximately  $-7 \times 10^{-5}$  in  $\Delta v$  in the 62% solution. Experimental checks have failed to show that there is any reason to suspect the original data and, as this point occurs near the middle of the concentration range, its importance cannot be minimized. However, the simplicity of equation (13) together with the fact that it yields a value of  $U_2/U_1$  which not only is independent of temperature, pressure, and concentration but also is the same as appears in the equations relating the attractive internal pressures and the optical absorption coefficients<sup>14</sup> to the compositions of the solutions, gives the equation a significance which justifies further study of its applicability.

The quantity  $-(\Delta_P \Phi_2 - \Delta_P V_2^0)$  which is a measure of the departures of the compressions (from 1 to 1000 bars) from the ideal law of mixtures is always positive in nitrobenzene-aniline solutions. This is an example of a case where the solutions are more compressible than the pure components and therein it differs from aniline-benzene or aniline-chlorobenzene mixtures. As far as we can tell from our data  $-(\Delta_P \Phi_2 - \Delta_P V_2^0)$  for each solution is independent of the temperature; and although  $-(\Delta_P \Phi_2 - \Delta_P V_2^0)$  may be calculated satisfactorily from a simple modification of equation (13), we may say that for interpolation purposes the equation  $-(\Delta_P \Phi_2 - \Delta_P V_2^0) = 0.1899 x_1$  gives a simple and adequate expression of the compression data at all temperatures.

The fact that when aniline and nitrobenzene are mixed only a small amount of heat is absorbed and an expansion occurs, together with the fact that this expansion diminishes as the temperature rises, is contributory evidence against compound formation in the ordinary sense of the term and the volume relations are such that on any simple hypothesis we must consider that if a compound were formed in these solutions, it would be destroyed by rise of pressure.

### Summary

We have measured the specific volumes at 25°, the thermal expansions at 10° intervals between 25 and 85°, and the compressions to various pressures up to 1000 bars at 25, 45, 65, and 85° of six solutions of aniline in nitrobenzene covering the whole range of concentration. These data have been represented by suitable equations from which the volumes, the thermal expansibilities

(13) R. E. Gibson, THIS JOURNAL, 69, 1521 (1937).

(14) Unpublished results obtained in this Laboratory.

and the compressibilities may be computed at any pressure and temperature within the range of our observations.

Our data enable us to compute  $(\partial P/\partial T)_V$ ,  $(\partial E/\partial V)_T$  and the volume changes on mixing for the various solutions, and to examine the variation of these quantities when the temperature is changed at constant volume. In the light of the results of this analysis, we suggest a correlation between the effect of temperature and of mixing on the molecular distribution in the liquids on the one hand and the internal pressures and the volume changes on mixing on the other.

The attractive pressures of the solutions were calculated in the same way as those of the pure liquids and were reproduced by the formula  $P_A = a'/V^{2.737}$ . An equation was developed whereby the constants  $a'$  for the solutions were represented as a function of the concentration in

terms of constants for the pure components and the generalized volume fractions  $y_1$  and  $y_2$  which contain an empirical constant,  $U_2/U_1$ .

Although the volume changes on mixing were well represented as functions of the ordinary volume fractions of the components by equations of a conventional type, we also examined several equations expressing this quantity in terms of significant properties of the solutions and the fractions  $y_1$  and  $y_2$ . These equations reproduced the volume changes on mixing fairly well. One of them had the form of the equations developed for regular solutions and another gave a value of the empirical constant,  $U_2/U_1$ , which was independent of temperature and pressure and was approximately equal to the corresponding constant obtained from the attractive pressure-composition equation.

WASHINGTON, D. C.

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[CONTRIBUTION FROM THE DEPARTMENT OF PHYSICAL CHEMISTRY, HARVARD MEDICAL SCHOOL]

## Serum Albumin. I. The Preparation and Properties of Crystalline Horse Serum Albumin of Constant Solubility

BY T. L. McMEEKIN

Since the crystallization of serum albumin by Gürber in 1894,<sup>1</sup> many investigators have doubted its homogeneity. Fractional crystallization from salt solutions has yielded products that vary widely in solubility,<sup>2</sup> chemical composition,<sup>3,4</sup> and dielectric properties.<sup>5</sup> Despite this evidence of inhomogeneity, the recognition and separation of constituent proteins has proved difficult. Perhaps the most important advance in its purification was the separation of carbohydrate-containing globulins<sup>6</sup> from the albumin fraction and the recognition that serum albumin does not contain carbohydrate.<sup>3,4</sup>

Serum albumin has been reported to be homogeneous in respect to size<sup>7</sup> and electrophoretic mobility.<sup>8</sup> Ultracentrifugal studies have shown it to have a molecular weight of 67,500,<sup>7</sup> independent of pH between 4 and 9. In solutions

more acid than pH 4 it was decomposed into smaller molecules, the process being found to be reversible.

This paper first describes our present procedure for the separation of the serum proteins and the preparation of crystalline carbohydrate-free serum albumin homogeneous with respect to electrophoresis at neutral reactions.<sup>9</sup> This highly purified serum albumin can be separated further into two crystalline fractions differing in chemical composition and electrophoretic mobility in acid but not in neutral solution.<sup>9</sup> The separation of serum albumin into two fractions is based on the discovery made in this Laboratory by J. D. Ferry,<sup>10</sup> that a salt-free concentrated solution of serum albumin crystallizes when brought to pH 4.0 with sulfuric acid.<sup>11</sup> The solubility of the

- (1) Gürber, *Würsburger physiol. med. Ges.*, **113** (1894).
- (2) Sørensen, *Compt. rend. trav. Lab. Carlsberg*, **18**, No. 5, 1 (1930).
- (3) Sørensen and Haugaard, *ibid.*, **19**, No. 12 (1933).
- (4) Hewitt, *Biochem. J.*, **30**, 2229 (1936).
- (5) Ferry and Oncley, *THIS JOURNAL*, **60**, 1123 (1938).
- (6) Hewitt, *Biochem. J.*, **32**, 26 (1938).
- (7) Svedberg and Sjogren, *THIS JOURNAL*, **50**, 3318 (1928); **52**, 2855 (1930).
- (8) Tiselius, *Biochem. J.*, **31**, 1464 (1937).

(9) I am indebted to Dr. John A. Luetscher, Jr., for these measurements. See also the second paper in this series, *THIS JOURNAL*, **61**, 2888 (1939).

(10) Reported by Ferry to the Division of Biological Chemistry at the 96th meeting of the American Chemical Society, Milwaukee, Wis., September 7, 1938.

(11) Piettre, *Compt. rend. Acad. Sci.*, **138**, 463 (1929), reports that among other proteins serum albumin crystallizes from aqueous solution following removal of globulin with acetone, and of acetone by evaporation over sulfuric acid *in vacuo*.