

SELF-ASSOCIATION OF ANTIPYRINE AND ITS DERIVATIVES IN AQUEOUS SOLUTIONS AT 40°C *via* OSMOMETRY

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The osmotic and activity coefficients of antipyrine, 4-methylantipyrine, 4-aminoantipyrine, and aminopyrine were determined in dilute aqueous solutions at 40°C using the vapour phase osmometric method. The osmotic data obtained were analyzed in terms of self-association of the compounds studied, based on several simplified association models treated by a numerical minimization method. The most probable type of stepwise self-association of the substances in aqueous solutions was found and the corresponding association constants were determined. The association is obviously accounted for by the "stacking" interactions of the pyrazolone rings as well as by hydrophobic and polar interactions.

Associations of organic molecules or ions in dilute aqueous solutions are being thoroughly studied in terms of the nature of mutual interactions of these particles and the specific part of the medium¹⁻⁶. The information on the formation of molecular associates in aqueous solutions is prerequisite for the study of interactions of bioactive substances with a biologic substrate and for the investigation of the so-called secondary structures of biomacromolecules themselves^{1,6-8}.

At present, NMR represents an efficient technique for the investigation of the mechanisms of formation and structures of molecular aggregates⁸⁻¹⁰. The determination of a more complex stoichiometry of association and of the corresponding equilibrium constants in aqueous solutions, however, encounters difficulties with that technique⁸⁻¹⁰. The equilibrium data are therefore found by other methods; in the case of substances sufficiently soluble in water, one can make use also of colligative methods, such as osmometry in the vapour phase^{1,2,11} used in this work. In the paper present the osmotic properties of antipyrine and its derivatives in aqueous solutions, determined by that technique, are reported. As the results indicated essential self-association of the substances dissolved, a numerical minimization procedure was set up, making it possible to determine — by means of appropriate association models — the association stoichiometry and the corresponding equilibrium constants from the osmotic data. The hitherto used ways of evaluation of association parameters from colligative data^{1,2,12,13} did not enable to describe the

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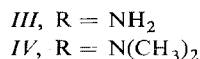
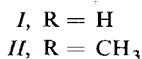
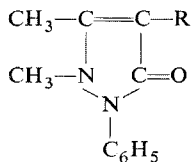
behaviour of the substances studied properly, or did not suit the character of the experimental data obtained.

Antipyrine and its derivatives were chosen for the osmotic study, thus following up our previous works, in which the heats of solution in water¹⁴ and thermodynamics of protonic dissociation^{15,16} were examined. Regarding their low basicity^{16,15}, antipyrine and its derivatives selected can be considered as nonelectrolytes for osmotic studies in aqueous solutions.

EXPERIMENTAL

Chemicals

Antipyrine (1,2-dihydro-1,5-dimethyl-2-phenyl-3*H*-pyrazol-3-one, *I*), 4-methylantipyrine (*II*), 4-aminoantipyrine (*III*), and aminopyrine (4-(dimethylamino)antipyrine, *IV*) were — with the exception of *II* (Hoechst) — of Czechoslovak origin, *purum* quality. The compounds were purified by recrystallization: *I* from ethanol, *II* from methanol, *II* and *IV* from benzene. Sodium chloride, reagent grade purity, was used without further purification. Aqueous solutions of required molality (mol/kg) were prepared from a weighed amount of dried substance (*II* over P₂O₅) and redistilled water.



Osmotic Measurements

The measurements were performed on an instrument Dampdruck-Osmometer (Knauer, Model 1970) at 40°C. Details are reported in the preceding paper¹⁷. In this work we applied a whole-metal two-termistor measurement probe; we did not use the recording equipment¹⁷ which appeared to be a source of inaccuracy. The calibration of the probe with aqueous solutions of sodium chloride was retained although the range of molalities was extended. The required osmotic coefficients of NaCl at 40°C were obtained by a recalculation from the activity coefficients (γ), modifying the method described^{18,19}.

The γ values published²⁰⁻²³ for NaCl solutions, molalities 0.0016–1.0 mol/kg (40°C), were expressed as a function of the molality, of the form $\ln \gamma = -A \sqrt{m}/(1 + B \sqrt{m}) + Cm + Dm^2 + \dots$. The theoretical coefficient A is 1.2016 (40°C)¹⁹; repeated calculations by the least squares method gave the values of the empirical coefficients $B = 1.4$ (set by trial), $C = 0.05497$, and $D = 0.02172$. The final calculation was performed with 18 selected values²⁰⁻²³ of $\ln \gamma$, which differed from the resultant course of the function by less than ± 0.0005 , calculated standard deviation ± 0.00022 . The standard deviation was minimum with the mentioned number of empirical coefficients. Osmotic coefficients (ϕ) of NaCl in aqueous solutions up to 1.0 mol/kg (40°C) can be

calculated using the above values according to the relation

$$\varphi = 1 - (A/B^3 m) [1 + B \sqrt{m} - 1/(1 + B \sqrt{m}) - 2 \ln(1 + B \sqrt{m})] + (1/2) C m + (2/3) D m^2. \quad (1)$$

The values calculated from Eq. (1) differed from those obtained, *e.g.*, by Lang¹⁹ (0.1–2.0 *m*) by less than 0.001. From the φ values we calculated the osmotic molalities \bar{m} of calibration NaCl solutions, defined as $\bar{m} = \nu \varphi m$, where ν is the number of ions in a "molecule" of the electrolyte and m is the stoichiometric molality of the latter. We used 14 calibration solutions of NaCl, 0.03–0.42 mol/kg; for the last of them ($\bar{m} = 0.7716$) the measured differences of the thermistor resistance ΔR of three probes employed lay in the range of 290–320 Ω . The dependences of the measured ΔR 's on \bar{m} of the NaCl calibration solutions were expressed as polynomials using the least squares method, the criterion of the appropriate degree of the polynomial was the minimum deviation of the ΔR values (± 0.7 to $\pm 1.0 \Omega$). For all the three probes used the calibration dependence $\Delta R = f(\bar{m})$ approached linearity, although in one case a polynomial of the third degree was the most suitable.

From the measured ΔR values for aqueous solutions of the studied substances *I*–*IV* at 40°C, the corresponding \bar{m} values were read from the tabulated calibration dependence $\Delta R = f(\bar{m})$. The results, representing averages of 4–5 independent measurements, are given in Table I. The lower number of determined \bar{m} 's for the compound *II* was due to an insufficient amount of the substance, in the case of the compound *IV* this was due to its low solubility — the 0.3 *m* solution is in fact oversaturated. Absolute standard deviations of the measured ΔR 's and thus of \bar{m} 's were in the region treated virtually independent of concentration. In the case of \bar{m} 's this was in average ± 0.002 – 0.003 mol/kg, the antipyrine solutions were worse measurable.

RESULTS

Osmotic coefficients were calculated from the experimental osmotic molalities (Table I) according to the relation given above, which for the studied non-electrolytes ($\nu = 1$) takes on the form

$$\bar{m} = \varphi m. \quad (2)$$

The experimental values of φ so obtained were expressed as a function of molalities of the compound in question, having the form of a polynomial as recommended^{1,2}:

$$\varphi = 1 + \sum_{i=1}^j a_i m^i. \quad (3)$$

The criterion of the appropriate degree of the polynomial, j (4–6), was the minimum standard deviation of the recalculated φ values. The empirical coefficients a_i found by the least squares method are given in Table II. The activity coefficients γ can be

TABLE I

Experimental Osmotic Molalities (\bar{m}) in Dependence on Stoichiometric Molality (m) (aqueous solutions, 40°C)

Compounds I and III $m(\bar{m}_I; \bar{m}_{III})$					
0.05 (0.047; 0.047)		0.45 (0.315; 0.288)		0.75 (0.441; 0.392)	
0.10 (0.089; 0.089 ₅)		0.48 (0.325; 0.301)		0.78 (0.454; 0.400)	
0.15 (0.128 ₅ ; 0.127)		0.51 (0.342; 0.314)		0.81 (0.465; 0.409)	
0.20 (0.164 ₅ ; 0.159)		0.54 (0.356; 0.323)		0.84 (0.471; 0.419)	
0.25 (0.198; 0.192)		0.57 (0.368; 0.337 ₅)		0.87 (0.484; 0.427 ₅)	
0.30 (0.230; 0.220)		0.60 (0.382; 0.345)		0.90 (0.496; 0.439)	
0.33 (0.246; 0.236)		0.63 (0.395; 0.352)		0.93 (0.510; 0.449)	
0.36 (0.261; 0.250)		0.66 (0.404; 0.365)		0.96 (0.520; 0.460)	
0.39 (0.279; 0.263)		0.69 (0.415 ₅ ; 0.377)		0.99 (0.532; 0.473)	
0.42 (0.298; 0.276)		0.72 (0.425; 0.383)		1.02 (0.543; 0.484)	
Compound II $m(\bar{m})$					
0.05 (0.043)	0.25 (0.162)	0.45 (0.239)	0.65 (0.283)	0.85 (0.316)	
0.10 (0.079)	0.30 (0.186)	0.50 (0.252)	0.70 (0.292)	0.90 (0.324)	
0.15 (0.110)	0.35 (0.206)	0.55 (0.264)	0.75 (0.299)	0.95 (0.330)	
0.20 (0.137)	0.40 (0.224)	0.60 (0.274)	0.80 (0.307)	1.00 (0.337)	
				1.05 (0.341 ₅)	
Compound IV $m(\bar{m})$					
0.04 (0.036 ₅)	0.12 (0.100)	0.20 (0.157 ₅)	0.27 (0.197)		
0.08 (0.069)	0.16 (0.130)	0.24 (0.180)	0.30 (0.214)		

TABLE II

Empirical Coefficients a_i of the Concentration Dependence of the Osmotic Coefficients (Eq. (3))

Compound	a_1	a_2	a_3	a_4	a_5	a_6
I	-1.3720	3.539	-7.745	9.912	-6.564	1.765
II	-3.143	12.941	-33.56	46.31	-31.78	8.562
III	-1.2239	1.381	-1.148	0.467	—	—
IV	-2.922	21.88	-87.70	121.9	—	—

then evaluated from the osmotic coefficients according to the relation²⁴

$$\ln \gamma = (\varphi - 1) + \int_0^m (\varphi - 1) d \ln m. \quad (4)$$

Substituting from Eq. (3) and integrating we obtain

$$\ln \gamma = \sum_{i=1}^j [(i + 1)/i] a_i m^i. \quad (5)$$

Eqs (3) and (5) yield statistically balanced values of φ and γ ; these are given for the compounds studied and the span of molalities 0.1–1.0 in Table III.

Evaluation of Association from Osmotic Data

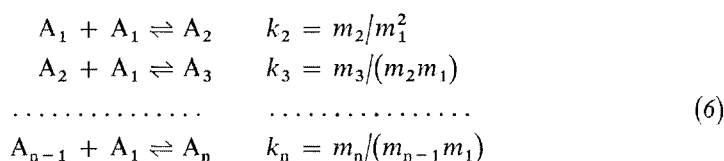
The determined osmotic molalities of the compounds studied (Table I) are regularly essentially lower than the corresponding stoichiometric molalities (or $\varphi < 1$, see Table III). As dilute solutions of nonelectrolytes are under study, this phenomenon indicates a considerable self-association of the compounds in question. This self-

TABLE III

Balanced Values of Osmotic (φ) and Activity (γ) Coefficients of Antipyrine and Its Derivatives (aqueous solutions, 40°C)

Molality	I		II		III		IV	
	φ	γ	φ	γ	φ	γ	φ	γ
0.1	0.891	0.794	0.786	0.623	0.890	0.798	0.851	0.699
0.2	0.819	0.669	0.685	0.469	0.802	0.658	0.784	0.579
0.3	0.763	0.582	0.620	0.382	0.730	0.557	0.713	0.486
0.4	0.716	0.515	0.561	0.320	0.670	0.482		
0.5	0.673	0.461	0.504	0.272	0.619	0.423		
0.6	0.635	0.416	0.455	0.236	0.575	0.376		
0.7	0.602	0.380	0.416	0.208	0.538	0.339		
0.8	0.574	0.350	0.386	0.186	0.508	0.308		
0.9	0.551	0.325	0.360	0.168	0.487	0.284		
1.0	0.536	0.304 ₅	0.334	0.153	0.476	0.266		

-association can generally proceed in a manner described by stepwise equilibria^{1,2,4}:



The symbols A_1, A_2, \dots, A_n designate the monomer, dimer, ..., n -mer of the dissolved compound, m_1, m_2, \dots, m_n the corresponding equilibrium molalities of those species, and k_2, k_3, \dots, k_n are the pertaining partial association constants. In dilute solutions of nonelectrolytes, osmotic molality can be considered as the sum of the equilibrium molalities m_i , and stoichiometric molality as the sum of the terms $i m_i$. Expressing the terms m_i ($i = 1$ to n) from the system of equations (6) and rearranging we obtain

$$\bar{m} = m_1 + \sum_{i=2}^n \left(\prod_{j=2}^i k_j \right) m_1^i \quad (7)$$

and

$$m = m_1 + \sum_{i=2}^n i \left(\prod_{j=2}^i k_j \right) m_1^i. \quad (8)$$

Differentiating Eq. (7) with respect to m_1 and dividing Eq. (8) by that term we find $d\bar{m}/dm_1 = m/m_1$, and by differentiating Eq. (2) we obtain $d\bar{m} = \varphi dm + m d\varphi$. We join these two equations and rearrange to obtain

$$d \ln(m_1/m) = (\varphi - 1) d \ln m + d\varphi. \quad (9)$$

Since for $m \rightarrow 0$, $\lim(m_1/m) = 1 = \lim \varphi$, we obtain by integration of Eq. (9) (in the limits from $m = 0$ to m) an expression, whose right side is identical with that of Eq. (4), hence $m_1/m = \gamma$. With regard to Eq. (5), the equilibrium molality of the monomer, m_1 , can be so evaluated from the expression

$$m_1 = (1/m) \exp \left\{ \sum_{i=1}^j [(i+1)/i] a_i m^i \right\}, \quad (10)$$

employing the coefficients a_i as given in Table II.

Eqs (6)–(10) represent fundamental relations for the evaluation of association of nonelectrolytes from experimental osmotic data^{1,2,12,13,25}, modified for the purposes of the present work. With regard to limited accuracy and to the concentration span of the experimental data, it is usually necessary to simplify the system (6) for

practical evaluation, which does not, however, affect the derivation and validity of Eqs (9) and (10). Usually only one or two mutually independent values of partial association constants are considered, which enables their evaluation by combining numerical and graphical methods^{1,2}. Simplified association models were used in this work, too, the corresponding association constants and the degree of the highest associate (n) were, however, evaluated purely by the numerical method, which allows — as compared with the graphical evaluation — a much higher variability of the models treated. The method consists in the minimization of the function

$$U = \sum_{i=1}^s (\bar{m}_{i,\text{obs.}} - \bar{m}_{i,\text{calc}})^2, \quad (11)$$

where $\bar{m}_{i,\text{obs}}$ are experimental (Table I) and $\bar{m}_{i,\text{calc}}$ the calculated values of osmotic molalities corresponding to a stoichiometric molality m , s is the number of experimental values. This approach is vindicated by successful application of minimization procedures in the study of complexes²⁷, micelle formation²⁶, etc. The above form of the minimization function, (11), was chosen with regard to the roughly equal accuracy of the experimental osmotic molalities throughout the whole region measured. For a mutual comparison of the various models, a quantity σ was defined by using the minimum U_{min} , the former representing in fact an estimate of the standard deviation of \bar{m}_{calc} from experimental osmotic molalities:

$$\sigma = [U_{\text{min}}/(s - p)]^{1/2}. \quad (12)$$

The symbol s has the same sense as in Eq. (11), p is the number of independent association constants (or the number of independent quantities by which they are mutually bound). In this work we confined ourselves to models with $p = 1-2$, which, however, characterize well enough the association equilibria of the nonelectrolytes studied.

Model 1. The formation of only one association species containing n molecules, i.e. $nA_1 \rightleftharpoons A_n$, $k = m_n/m_1^n$. According to Eq. (7), $\bar{m}_{\text{calc}} = m_1 + km_1^n$ is substituted in the minimization function (11). The equilibrium molalities of the monomer, m_1 , are calculated from Eq. (10); those values m , at which the various \bar{m}_{obs} 's were measured, are used in the calculations (Table I). Values of k and n are substituted by trial until U_{min} is found; $p = 1$ in Eq. (12).

In contrast to Model 1, the other models involve the stepwise association described by the system (6).

Model 2. The identity of all the partial association constants of the system (6), i.e. $k_2 = k_3 = \dots = k_n = k$ is assumed. Substituting in Eq. (7) and rearranging we

obtain for the model osmotic molality $\bar{m}_{\text{calc}} = m_1[(km_1)^n - 1]/(km_1 - 1)$. The following procedure is identical with that for Model 1, also $p = 1$.

Model 3. In the system (6) an independent dimerization constant k_2 and equality of all the other partial association constants, $k_3 = k_4 = \dots = k_n = k$, is assumed. Substituting in Eq. (7) we obtain $\bar{m}_{\text{calc}} = m_1 + k_2 m_1^2 [(km_1)^{n-1} - 1]/(km_1 - 1)$. Further proceeded as with Model 1; the values of k_2 , k , and n are adjusted by trial, $p = 2$.

Model 4. In the system (6) the dimerization constant is independent, the other partial constants are considered as related through $k_i = q/i$, where q is an empirical coefficient. Thus the values of the partial association constants k_3 to k_n decrease regularly with increasing i , and $k_\infty = 0$. For this model we obtain from Eq. (7) $\bar{m}_{\text{calc}} = m_1 + 2k_2 \sum_{i=2}^n (q^{i-2}/i!) m_1^i$. Further procedure as with Model 1, the k_2 , q , and n values are adjusted by trial, $p = 2$.

Model 5. Again the k_2 value is independent, the other partial association constants are interrelated by $k_i = q(i-2)/(i-1)$, where q is again an empirical coefficient. So, in contrast to the preceding model, the values k_3 to k_n increase slightly, with $k_\infty = q$. Substitution in Eq. (7) yields $\bar{m}_{\text{calc}} = m_1 + k_2 \sum_{i=2}^n [1/(i-1)] q^{i-2} m_1^i$. Further procedure is identical with that for Model 4.

During the seeking for the minimum of the function U (Eq. (11)) the parameters k_2 , k and q were varied with the step lower than 1%, n was adjusted as appropriate in the range of 2–15. An approximate estimate of the starting value of the independent dimerization constant k_2 (Models 3–5) can be obtained graphically^{1,2}. The found values of the parameters for the various models corresponding to U_{min} are summarized in Table IV, along with the values σ (Eq. (12)), which are more illustrative than the U_{min} 's. The hitherto performed analyses^{1,2} of real experimental osmotic data using a combination of numerical and graphical methods were usually restricted to the association models of the type 1–3. The evaluation of the parameters of the more complex Models 4 and 5 *etc.* usually required spectrophotometric, extraction, and other data^{5,3}. The minimization method described makes it possible to evaluate from colligative data the parameters of these and other association models of non-electrolytes by a unique procedure. The option of a different model is reflected only by the expression for \bar{m}_{calc} .

The calculations were performed on a computer CDC 3300 of the Research Computer Centre of the UNO Programme, Bratislava. The programmes required were set up in the language Fortran.

DISCUSSION

The association models described were chosen so that the mutual confrontation of their results (Table IV) could yield the best image of the self-association of the studied nonelectrolytes in aqueous solutions. A detailed analysis should be carried out especially for the compounds *I–III*, which cover a broader concentration range. As the least suitable for those compounds appeared the association models 1 and 2; the corresponding σ values (Eq. (12), Table IV) exceeded here the estimates of the average standard deviations of the experimental \bar{m} 's ($\pm 0.002–0.003$). The self-association of the compounds *I–III* is obviously not restricted to the formation of a single associate species (Model 1), but proceeds according to the step equilibria (6) with the formation of the set of multimers (Models 2–5). The partial association constants k_2, k_3, \dots, k_n cannot, however, be considered equal to each other, although such a simplifying assumption is not infrequently employed^{1,2,10}. Models 3–5 therefore involve an independent value k_2 and mutually opposite trends of the constants k_3, k_4, \dots, k_n . As the most appropriate, Model 5 appeared for the compounds *I–III*, with an increase in the sequence of association constants ($k_\infty = 2k_3$). The optimum selection of the Model 5 follows from the lowest σ values (Table IV); as a complementary criterion we followed also the uniformity of distribution of

TABLE IV
Parameters of the Association Models Calculated from Osmotic Data of Antipyrine and Its Derivatives

Model	(<i>n</i>)	k_2	k, q^a	($\pm \sigma \cdot 10^3$)	Model	(<i>n</i>)	k_2	k, q^a	($\pm \sigma \cdot 10^3$)
Compound I					Compound II				
1	(3)		8.30	(5.83)	1	(3)	48.3		(4.88)
2	(≥ 12)		1.40	(3.21)	2	(≥ 15)		3.47	(4.80)
3	(≥ 12)	1.25	1.63	(1.80)	3	(≥ 15)	2.76	4.16	(2.50)
4	(≥ 8)	1.14	6.50*	(2.01)	4	(≥ 10)	2.24	18.85*	(3.25)
5	(≥ 13)	1.36	2.45*	(1.68)	5	(≥ 15)	3.26	5.74*	(1.80)
Compound III					Compound IV				
1	(3)		10.75	(3.69)	1, 2	(2)		3.14	(0.761)
2	(≥ 12)		1.59	(8.07)	3	(≥ 5)	2.90	0.58	(0.559)
3	(≥ 13)	1.11	2.35	(1.41)	4	(≥ 5)	2.86	2.08*	(0.554)
4	(≥ 10)	0.90	10.60*	(1.85)	5	(≥ 6)	2.87	1.27*	(0.549)
5	(11)	1.26	3.38*	(1.35)					

^a The q values are marked by an asterisk.

the differences $\bar{m}_{\text{obs}} - \bar{m}_{\text{calc}}$ in dependence on stoichiometric molality. Some irregularity was observed only for the derivative *II*.

Model 3, used owing to its simplicity in other osmotic studies² and recommended for solutions of nonelectrolytes⁵, did not prove to be satisfactory enough in the case of compounds *I–III*. We tested also a model with interdependence of all partial association constants. It confirmed the trend of increase in the sequence of the constants, but it was not better than Model 5. Thus for an approximate calculation of the equilibrium molalities of the various multimers of the compounds *I–III* according to Eqs (6) and (10) the values of association constants corresponding to Model 5 can be recommended. As indicated by results with Models 2–5, the magnitude of n , *i.e.* the number of monomers in the highest multimer, is not appreciably restricted. The σ values usually decrease with increasing n up to the limit value shown in Table IV. Model 5 appears as the most suitable for the low-soluble derivative *IV*, too. Regarding, however, the low values of the constants $k_3 - k_n$ (Models 3–5) and a narrow accessible concentration range, one could explain the osmotic properties of the derivative *IV* quite well in terms of the dimer formation solely (Models 1 and 2, Table IV).

Mukerjee and Ghosh⁴ pointed out two limit types of self-association of organic substances in aqueous solutions. The first type is formation of dimers, which agglomerate to form big micelles. The second type is characterized by a successive formation of a numerous set of multimers A_2, A_3, \dots, A_n . In fact, this second type covers also the substances studied, possibly with the exception of the derivative *IV*. A stepwise formation of multimers is usually accounted for by the so-called "stacking" or dispersion π - π interactions between planar heteroaromatic molecules or dye ions^{3,5}, nucleic acids and bases, and other biologically significant substances^{9,10}. A characteristic feature of the stacking interactions is cooperativeness, linking of a monomer to a dimer is easier than the formation of the dimer itself^{6,2}. In the case of validity of Model 3, recommended for solutions of nonelectrolytes⁵, the cooperativeness shows up in the ratio k_3/k_2 being greater than 1. So is it in the case of the compounds *I–III* too (Table IV, Model 3). Since, however, for the substances under study the Model 5, characterized by an increase of the higher association constants, proved to be more appropriate, we suggest for this model as an analogous criterion of cooperativeness the value of the ratio $k_\infty/k_2 = q/k_2$. These ratios are (in descending order) 2.68 (*III*), 1.80 (*I*), 1.76 (*II*), and 0.44 (*IV*). The stacking interactions obviously contribute essentially to the stabilization of the associates of the compounds *I–III*, whereas the noncooperativeness of association of *IV* indicates a different mechanism.

The dependence of the k_∞/k_2 ratios on the type of substituent suggests that self-association of the compounds *I–III* is conditioned by mutual stacking interactions of their pyrazolone rings (Scheme 1) possessing aromatic character^{28,29}. In the stabilization of the associates can take part also hydrophobic interactions and electrostatic contributions, resulting from the high polarity of the CO group^{28–30}.

Immediate intermolecular hydrogen bonds can be ruled out, regarding the lack of a suitable H-donor. The bulk hydrophobic substituent $N(\text{CH}_3)_2$ probably hinders the stacking interaction of the pyrazolone rings, the dimerization of the derivative *IV* is obviously caused first of all by a hydrophobic interaction. In our recent work¹⁴ we found considerable negative heats of dilution of aqueous solutions of antipyrine and its derivatives (*I-IV*). This implies that the dissociation of molecular multimers, along with a higher hydrophobic and polar interaction of lower associates or monomers with water, is in general an exothermic process. As the heats of dilution¹⁴ were determined at a different temperature (25°C), the corresponding thermodynamic functions could not be evaluated with accuracy, but the positive values of the heats of association themselves point out the contribution of the aqueous medium to self-association of antipyrine and its derivatives occurring through hydrophobic interactions^{7,31,32}.

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