249. Comment on the Estimation of the Dimerisation Constant of Caffeine

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Summary

Reasons for the low reliability of the ^{1}H-NMR estimation of the dimerisation constant of caffeine in D_2O are discussed. The main reasons are *1*) strong influence of small variations in the monomer shifts δ_0 which are to be determined by extrapolation, and *2*) disturbance by the formation of higher oligomers. The dilemma ist that attempts to minimize errors from (*1*) increase the influence of (*2*) and *vice versa*.

Horman & Dreux [1] estimated the dimerisation constant K^D of caffeine in D_2O from ^{1}H-NMR shifts δ_i in a series of caffeine dissolutions varying in caffeine concentration. (There seems to exist a long-living error in the formula of caffeine: omission of one central bond.) For each of the four proton signals of caffeine three parameters were calculated simultaneously by a linearisation procedure: K^D, the chemical shifts δ_0 of the monomeric caffeine, and the chemical shifts δ_∞ of the dimeric caffeine. This is a convenient solution for the problem to estimate δ_0 by extrapolation.

We were engaged with the same investigation as was briefly mentioned in a recent paper [2]. We used essentially the same technique *(Bruker HX-90)* but did not publish our results because we considered any parameters computed from shift measurements of this system to be not very reliable. This is partly evident even from the results of *Horman & Dreux* [1], particularly in comparison to the very good accordance of complex parameters for binary heterocomplexes AB (*e.g.* caffeine/benzene) which can be obtained from different A signals and from different experimental series [3] [4]. *Horman & Dreux* mentioned that many anomalous results for AB complexes are known and that the results are often approximate. However, provided that the experimental conditions have been optimum, probably many of these anomalous results can be transferred into 'normal' (*i.e.* self consistent) results by performing the AUS correction [5], *i.e.* the correction of the additional unspecific shielding which is exerted on the A protons by B[1]). Remaining anomalies may be caused by the formation of higher complexes, *cf. e.g.* [6][2]).

[1]) Compare *e.g.* former results cited in [3] [4] with our results in [3].

[2]) Our unpublished results (*H.-O. Strumm & H. Stamm)* confirm that the picryl-acetone/benzene system cannot be described by the 1:1 model.

As far as comparisons can be made our own results for the caffeine dimerisation correspond more or less with the results of the authors (temperature, K^D in l/mol; $\delta_o - \delta_\infty$ in Hz measured at 90 MHz and calculated for 80 MHz): 5°: CH_3–N(1) 7.2; 34; CH_3–N(3) 8.6; 35; CH_3–N(7) 7.8; 25; 27°: CH_3–N(1) 6.5; 24; CH_3–N(3) 7.6; 28; CH_3–N(7) 7.8; 16; 80°: CH_3–N(1) 1.7; 22; CH_3–N(3) 1.7; 22; CH_3–N(7) 1.3; 12. These parameters were obtained by using known computation methods [7] on the basis of shift measurements *vs.* internal TSP. Since it is known [8] that *e.g.* the TSP signal is shifted nonlinearly by increasing purine concentrations, we also employed an external reference (cylindrical or spherical insert) without recognizable improvement in the computed parameters. Many further variations in experimental conditions or treatment of the data were not encouraging, and we can fully confirm the finding of *Horman & Dreux* that the computed K^D is remarkably sensitive to small variations in the monomer shift δ_o. However, the statement that the precision of the necessary extrapolation is favoured by employing a wide enough concentration range does not hold for this particular system. *Guttman & Higuchi* [9] concluded from their work that caffeine in water not only forms dimers but also tetramers. Their elegant treatment of simple partition data seems to be basically sound and gains plausibility from the finding that 1-ethyltheobromine forms no tetramer. Clearly, stacking of the dimer of 1-ethyltheobromine is prevented by the ethyl group which is hindered from taking on a coplanar conformation by the two neighbouring oxygen atoms. Further, K^D always has the same order of magnitude as the equilibrium constant K^T for the formation of one tetramer from two dimers.

Obviously, there is a dilemma in that attempts to decrease the influence of the tetramerisation (*e.g.* by using very low concentrations only) makes the extrapolation (*i.e.* the estimation of δ_o) uncertain and *vice versa*. So, for caffeine one has to expect serious difficulties in any trial to estimate reliable association constants from ^{1}H-NMR data because the results are not only subject to statistical and extrapolation errors but also to systematic errors.

Guttman & Higuchi [9] estimated $K^D = 11.7$ l/mol and 1750 l^3/mol^3 for the equilibrium between monomers and tetramers ($K^T \times K^D \times K^D$) at 30°. From the reported osmotic coefficient 0.65 of a 0.1M aqueous caffeine dissolution at 25° [10] and under the assumption that dimerisation is the sole equilibrium $K^D = 39$ l/mol is calculated. Assuming an incertitude of ±0.05 in the osmotic coefficient leads to the limiting values 19 and 100 l/mol. On the other hand, calculation on the basis of the two *Guttman-Higuchi* constants at 30° yields an osmotic coefficient 0.655 for a 0.1M caffeine dissolution.

The direct and computationally simple method described by *Horman & Dreux* may be valuable for pure dimerisations but has its limits when self association proceeds beyond the dimer stage. This is a general problem for any treatment of shift data.

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