

Abstract

G. Graziano claims that solubility differences can be rationalized by the reversible work needed to create a cavity in the solvent of the size of the solute. This quantity is closely related to the solvent accessible surface area, which is greater in the case of methanol than in water, making it thus more soluble. In his comment, G. Graziano stated that these properties are responsible of the huge difference between the solubilities of methanol and water. Moreover G. Graziano claims that the differences found in our original work (Henaio, et al. PCCP, 18, 23006 (2016)) are “small” and unable to explain that differences in solubility. We show in this reply to this comment that the increase of the solvent accessible surface area, and thus the work required for cavity creation, is insufficient to fully account for the increase of indole solubility in water by the addition of very small quantities of methanol. In other words, as stated in the original paper, methanol is *actively changing* the solvation shell and not just passively increasing the solvent accessible surface area. Secondly, we also compare the distance distribution functions, in an alternative way to that described in our original paper, describing the methanol and water solvation of indole to reinforce the fact that the differences in these functions are quantitatively greater than those found by Graziano. As a result of these additional analyses, we conclude that, our work on the solvation differences of indole in water and methanol successfully captures differences in the solvation shells of both solvents around indole. While we do agree that more work should be done on this field, we think that perhaps our results could be added to the calculations suggested by G. Graziano to lead to a more full description of the solubility.

Solubility is a paramount problem in many fields. One of the difficulties in understanding solubility is that experimental measures of this phenomenon are, necessarily, conducted on a macroscopic scale; yet the details by which this occurs on the atomic level are not well understood. Trying to couple a microscopic understanding of a bulk phenomenon is inherently difficult as a macroscopic measurement contains changes to the ensemble of molecules and the details of what happens to individual molecules within this ensemble is difficult to access by experimental measures. A theory which could predict solubility assuming a minimum number of parameters, but that still kept the fundamental microscopic details of molecules is, in fact, of the greatest importance.

It has been proposed by Graziano in a comment on our recent publication entitled *On the positional and orientational order of water and methanol around indole: a study on the microscopic origin of solubility* and in other works, that the reversible work needed to create a cavity of the size of solute in the solvent is the primary driving force in determining the solubility of a molecule. The magnitude of this work is related to the solvent accessible surface area (SASA), calculated by Graziano, after assuming spherical or spherocylindrical shapes of molecules. SASA is assumed to be a smooth surface containing the center of the solvent molecules surrounding the solute within the first hydration shell. While this approach does not

explain which level of approximation is necessary to assume the molecular shape, it is clear that the surface area increases for the same solute as the size of solvent molecules increase. To compare with our recent publication (Henao, et al. PCCP, 18, 23006 (2016)) Graziano assumed that tryptophan is a reasonable model for indole, and that tryptophan could be considered to have a spherical or a spherocylindrical (oblate or prolate) shape where all of these shapes share the same van der Waals volume. Indeed, following the comment from Graziano, the exact shape of indole is not important, since SASA will *always* increase by increasing the radius of the solvent. For this reason, the bigger the solvent molecule, the higher the solubility. In comparison to our work, Graziano argues that since the radius assigned to methanol is greater than that of water, the solubility of indole in methanol is greater. We believe that this assessment is incorrect and that solute-solvent interactions cannot be treated in this manner, as it is much too simplistic to explain the solubility of all molecules as it does not always hold true. Indeed, as an example, Graziano in a previous work has shown that urea and thiourea (both with almost the same van der Waals volume) molecular interactions can completely change the predictive solubility model.

The solubility of indole in water is substantially increased by the addition of small quantities of methanol to the solution. As stated in our original paper *“the addition of only 30 methanol molecules per indole, increases the water solubility 3000 times”*. If the work required for cavity creation is the main quantity for describing the solubility of indole in water and methanol, it would be expected a drastic increase in the SASA of the indole-water system when a small amount of methanol molecules is added. In other words, if this predication where true: SASA of indole in water should increase to values close to that of the SASA of indole in methanol at small proportions of methanol in water (X_{methanol}). Moreover, it should not follow a smooth trend between the two binary systems. In order to check this prediction we have calculated SASA of the system indole for different concentrations of methanol in water. SASA calculations were performed assuming a spherical shape for both the solute (indole) and the solvents water and methanol, being the radius for all the species the same as those in the comment of Graziano.

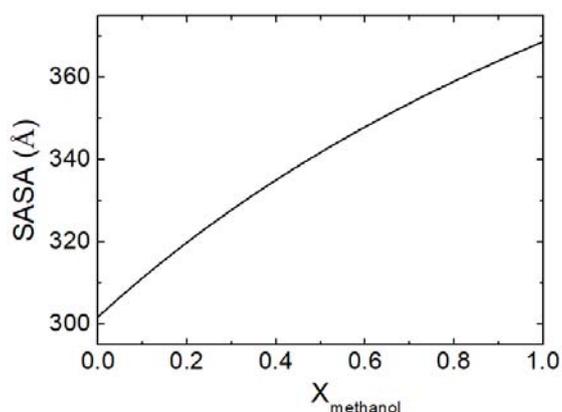


Fig.1.- Variation of the solvent accessible surface area for a mixture of water and methanol around Indole, assuming the parameters used by G. Graziano. For $X_{\text{methanol}}=0$ we obtain the binary mixture indole-water, while for $X_{\text{methanol}}=1$ the mixture Indole-methanol is recovered.

From this calculation (as shown in Fig. 1) the SASA varies continuously with increasing methanol concentrations, and, importantly, there is no evidence of a step-function change at low methanol concentrations, which would occur if the method described by Graziano were able to describe the solubility in our system. We conclude thus that the approach of Graziano is not able to capture the mechanism through which solubility increases for indole in methanol water systems. In other words, the solvent is not playing a passive role (as assumed by Graziano) but actively changing indole solvation. This, in fact, was one of the primary conclusions from our previous work: that methanol is *actively changing solvation* by expelling the water molecules in the first hydration shell surrounding indole .

Additionally, Graziano states that the changes reported in our work between the solvation of indole by methanol or water are so small that they cannot capture the large differences in solubility between the two systems. On the other hand, the calculations of Graziano find a maximum difference of about 20%. The differences reported in our publication are not small as suggested by Graziano suggests. In order to reiterate this we have produced distance distribution $g(r)$ s to show the difference between solvation of both methanol and water around indole from the data reported in our previous work.

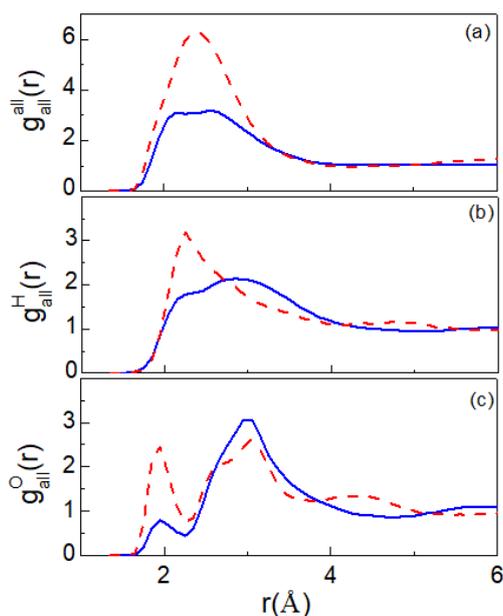


Figure 2.- Distance distribution functions (DDF) for the binary systems indole-water (blue, solid line) and indole-methanol (red, dashed line). Panel (a) shows the surface-surface DDF, panel (b) the surface-hydrogen DDF and panel (c) the surface-oxygen DDF.

Figure 2 shows a direct comparison of the distance distribution functions (DDF) found in our previous work. Three different DDF's are shown: one between the surface of solute and solvent (panel a), and two from the distances between the surface of indole and hydrogen or oxygen atoms (panels b and c) of water (blue) or methanol (red). A direct inspection of the figure makes clear that differences between indole in methanol and water are not small. The DDF's describing the two systems differs both in shape and magnitude. In order to quantify

this, the difference between the indole-solvent surface-surface DDF for water and methanol is close to 50% for the nearest neighbor interactions (low r values in Fig. 2), greater than those found by Graziano. Moreover, as stated in our original paper, the differences on the surface-oxygen clearly show the presence of a peak that is non-existent in the water-indole system. As described in our original paper, this additional peak is related to a solvation shell present between indole and methanol that is not present in the indole-water mixture. This is due to a difference in molecular orientation of methanol compared with water around the indole molecules, as seen in Figure 5 (c) of the original paper. This figure also shows that the two-dimensional distribution functions related to water and methanol $g(\psi^{\text{ori}}, \cos(\theta_{\text{ori}}))$ are completely different, the one related to methanol showing one spot, and the one of water showing two spots (see our original work for details). Finally, differences were also found in the calculated entropies in our work (Figures 6 and 8 of the original paper), where, again, both the shape and magnitude of the curves are substantially different between the indole-methanol and indole-water systems and differ by up to 70% at some points. Therefore, we think that differences in our case are quite large rather than being 'far too small' as claimed in the comment by Graziano.

In conclusion we think that a theory including molecular interaction could be of great interest for the scientific community. While Graziano's approach may be able to capture the steric contribution to solubility, at its present stage, this approach fails to adequately describe solubilities in systems where molecular interactions play a significant role, such as occurs for indole in methanol-water solutions. We believe that the differences found in our work (which are certainly not small) suggest that the solvent has an active role in solubility and hope that this information can be included in future investigations to augment calculations on the reversible work for cavity creation. This would allow for the thermodynamics of solubility to be assessed in a way that captures the important contribution of the molecular interactions in solutions.