

An X-ray and neutron scattering study of the equilibrium between trimethylamine N-oxide and urea in aqueous solution

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Supporting Material

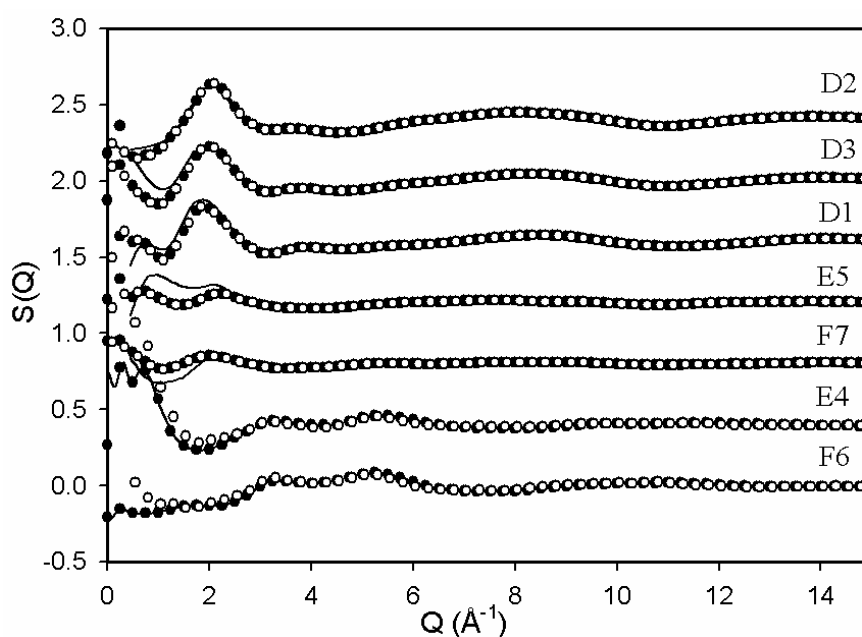


FIGURE S1: Comparison of the results of the simulations with the large (*solid circles*) and small (*open circles*) boxes with the experimental data (*full line*) for the TMAO-urea 1:1 solutions. Successive sets of curves have been displaced by 0.5 along the ordinate for better visualization. The letter code refers to the solution compositions given in (1).

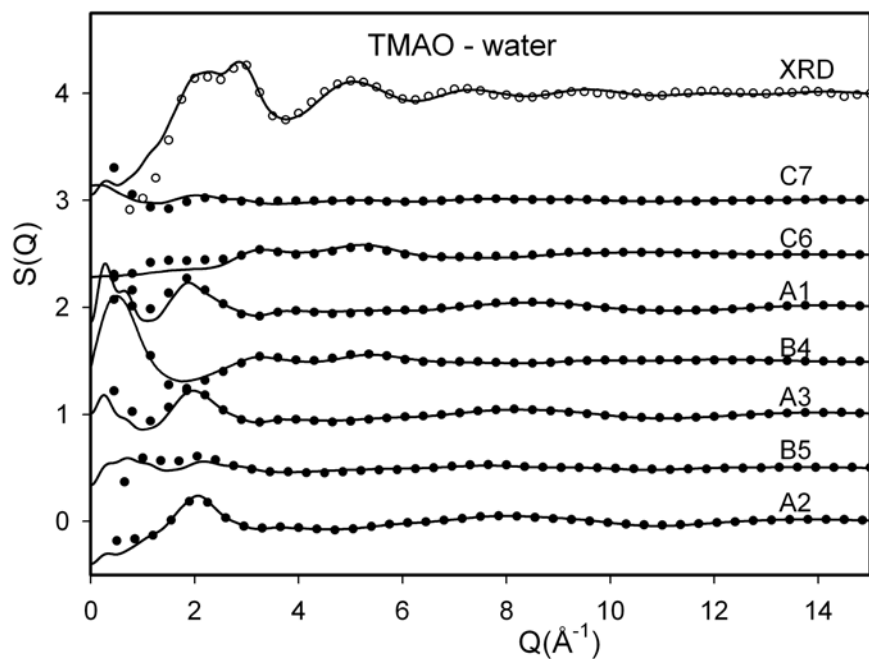


FIGURE S2: Structure factors $S(Q)$ for 2.5 M solutions of TMAO obtained by neutron and X-ray scattering (For the experimental neutron data 1:7 points (*solid circles*) is shown, for the X-ray data (*open circles*) 1:5) and EPSR fits (*solid line*). The statistical error on the experimental data is smaller than the size of the symbols. Successive sets of neutron scattering curves have been displaced along the ordinate by 0.5 for better visualization. The X-ray scattering curve was displaced by 4 units. These fits should be compared with those of Figure 1 (top) in (1) where the composition of the samples is also given in Table 1.

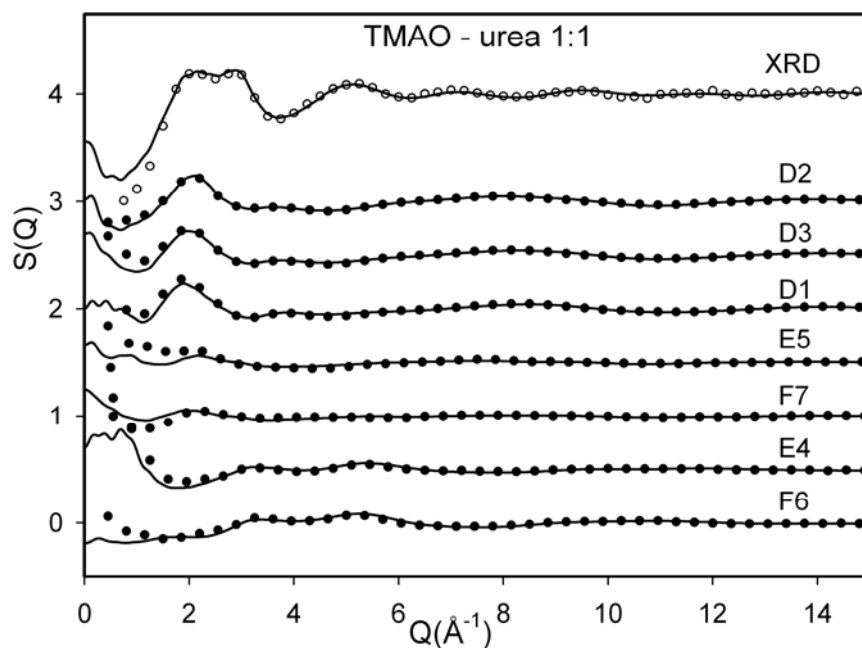


FIGURE S3: Structure factors $S(Q)$ for solutions containing 2.5M TMAO and 2.5M urea obtained by neutron and X-ray scattering (For the neutron data (*solid circles*) 1:7 points is shown, for the X-ray data (*open circles*) 1:5) and EPSR fits (*solid line*). The statistical error on the experimental data is smaller than the size of the symbols. Successive sets of neutron scattering curves have been displaced along the ordinate by 0.5 for better visualization. The X-ray scattering curve was displaced by 4 units. These fits should be compared with those of Figure 1 (bottom) in (1) where the composition of the samples is also given in Table 1.

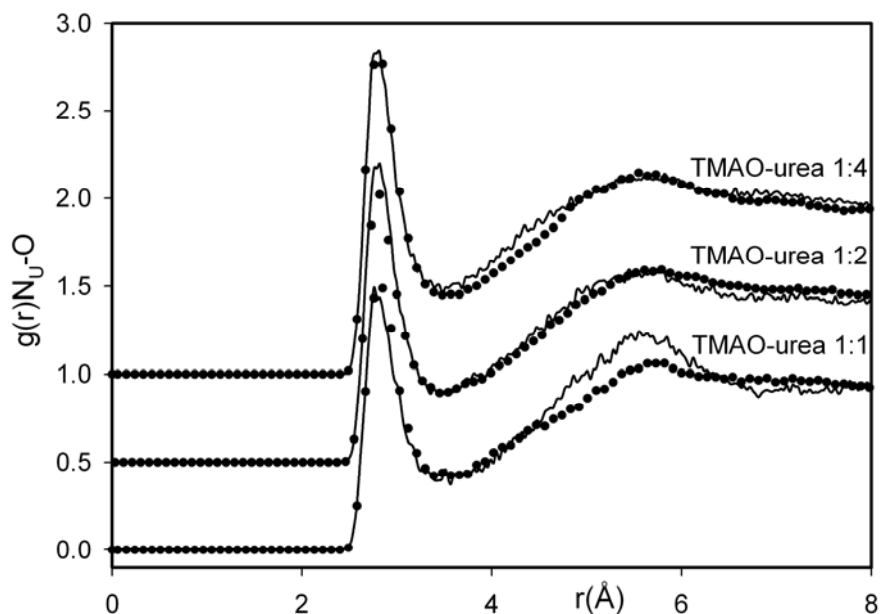


FIGURE S4: $g(r)_{NU-O}$ obtained from the neutron data only (*solid circles*) and from the combined X-ray and neutron data (*full line*). The curves have been displaced by 0.5 along the ordinate for better visualization.

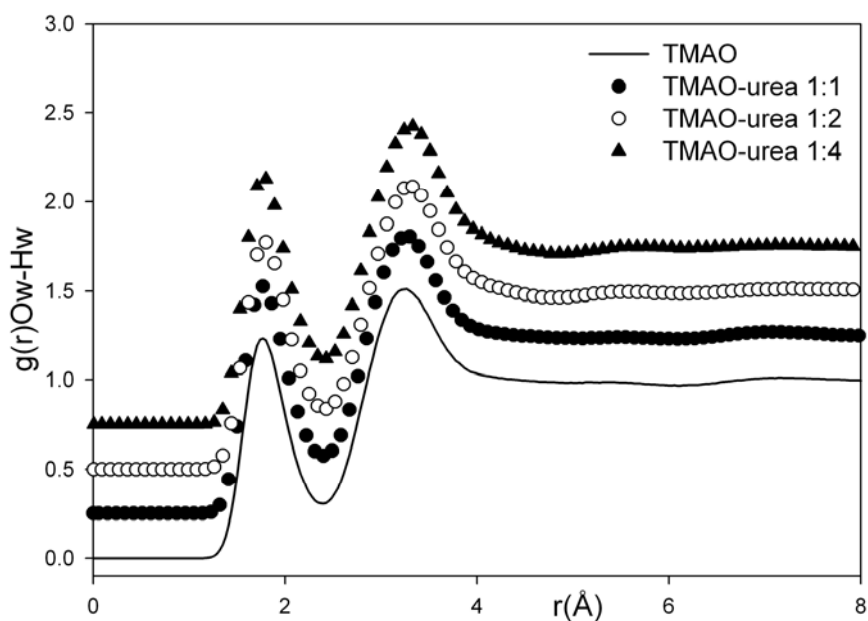


FIGURE S5: The first maximum of $g(r)_{Ow-Hw}$ occurs at 1.77 Å as in water rather than at 1.75 Å when only the neutron data are used. The position of the second maximum shifts from 3.25 Å in water to 3.27 (3.24) Å in TMAO-water, 3.24 (3.27) Å for TMAO-urea 1:1, 3.30 (3.31) Å for TMAO:urea 1:2 and 3.33 (3.32) Å for TMAO:urea 1:4. The values between brackets correspond to the refinement with the neutron data only.

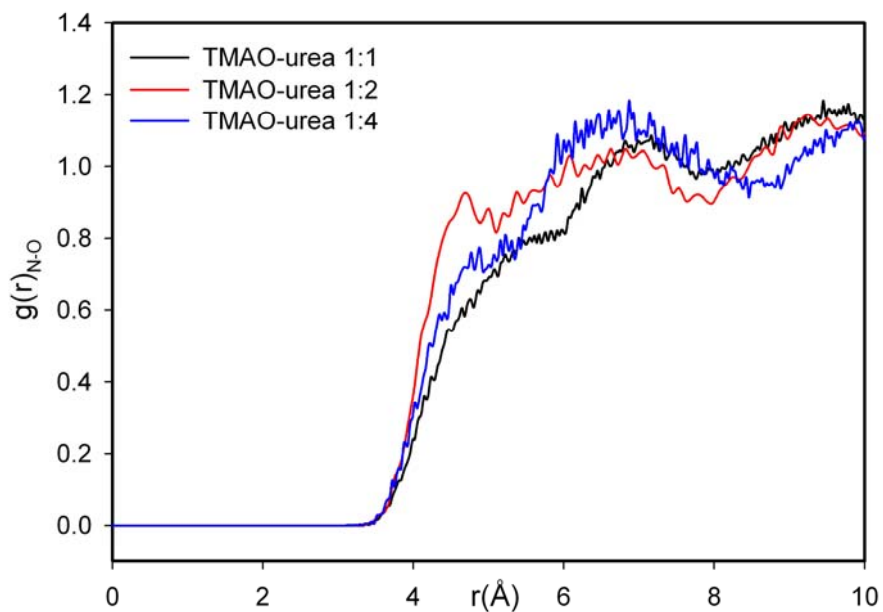


FIGURE S6: $g(r)_{N-O}$. Note that the N-O distances are not sharply defined in contrast with the OU-NU distances in Fig. S8.

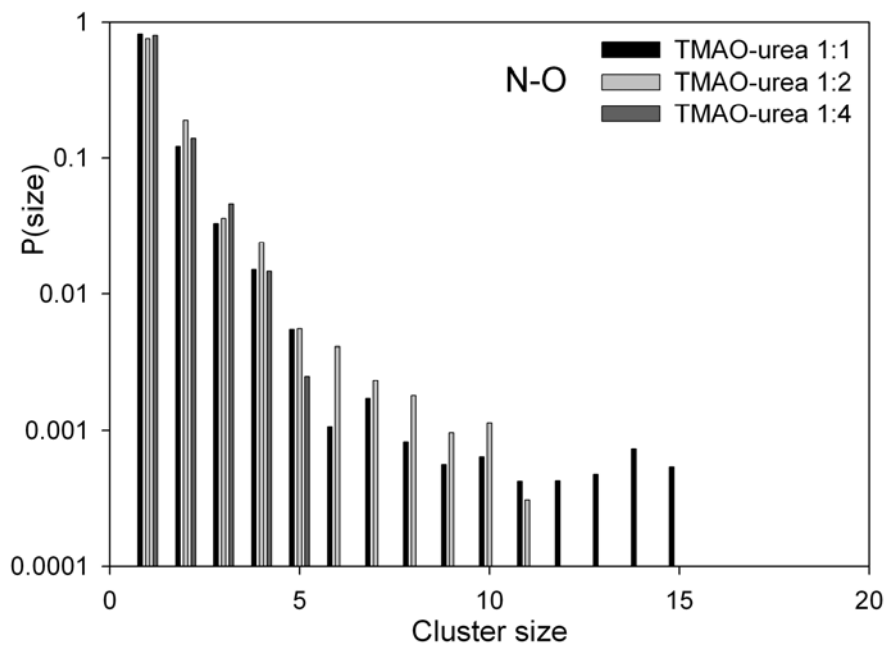


FIGURE S7: Size distribution of TMAO-TMAO clusters obtained with a cutoff distance of 5.4 Å. There is no sign of significant TMAO-TMAO association.

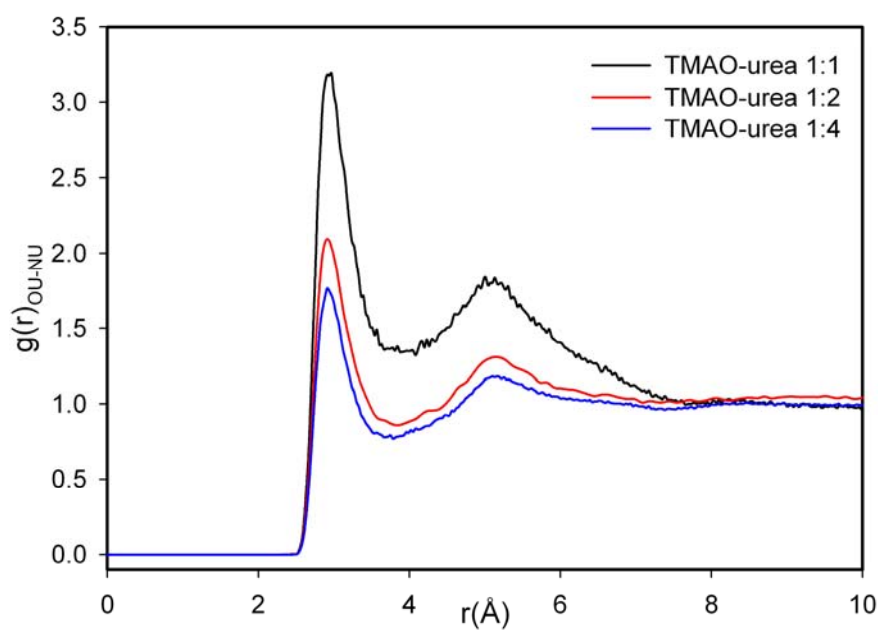


FIGURE S8: The rather sharp first coordination peak in the $g(r)_{OU-NU}$ suggests that well defined structures are formed by urea association.

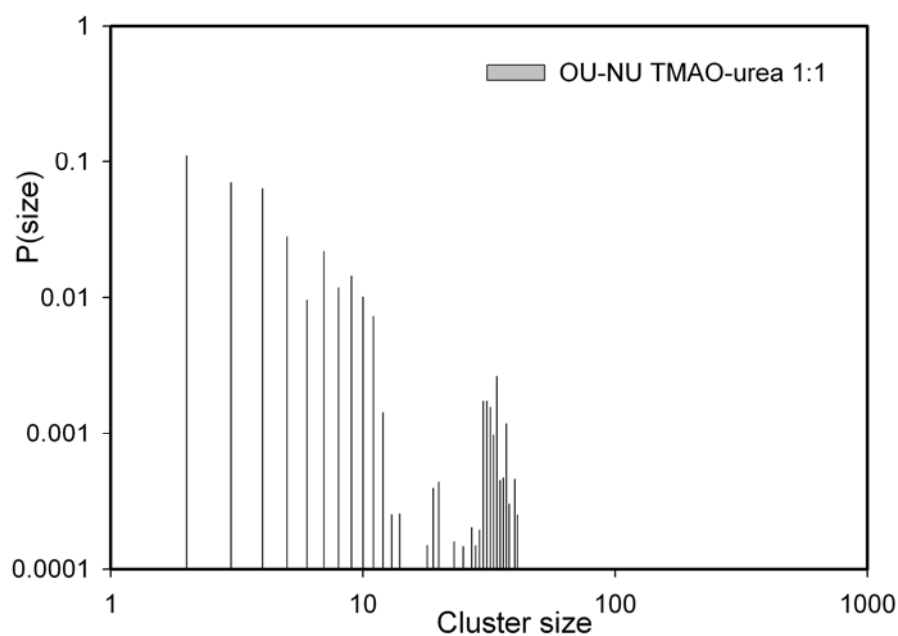


FIGURE S9: Size distribution of clusters of urea molecules with N_U-O_U distances in the range 1.0-3.8 Å for the TMAO:urea 1:1 solution. Note the logarithmic scales.

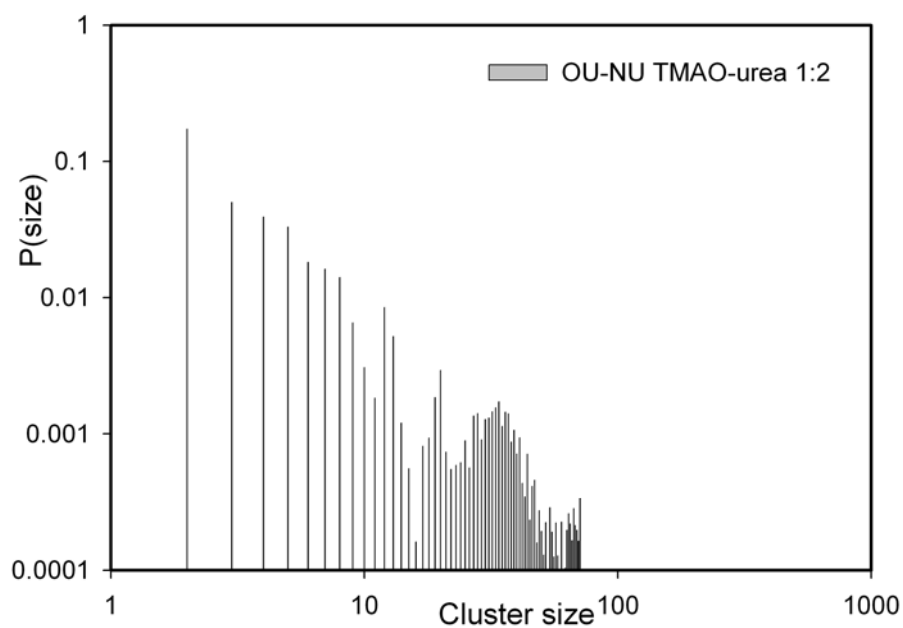


FIGURE S10: Size distribution of clusters of urea molecules with N_U-O_U distances in the range 1.0-3.8 Å for the TMAO:urea 1:2 solution. Note the logarithmic scales.

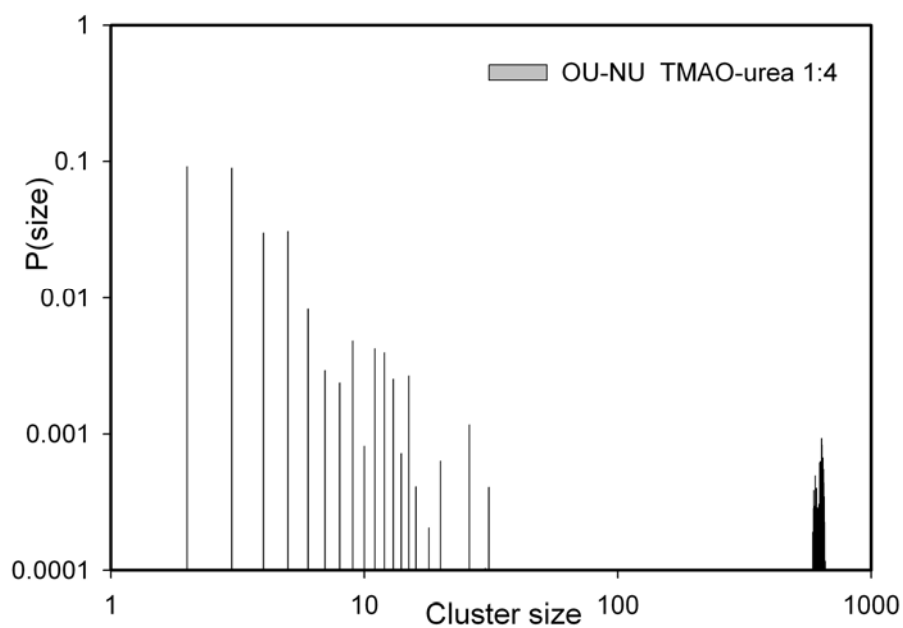


FIGURE S11: Size distribution of clusters of urea molecules with N_U-O_U distances in the range 1.0-3.8 Å for the TMAO-urea 1:4 solution. In contrast with the two other solutions this one is characterized by a few very large clusters. Note the logarithmic scales.

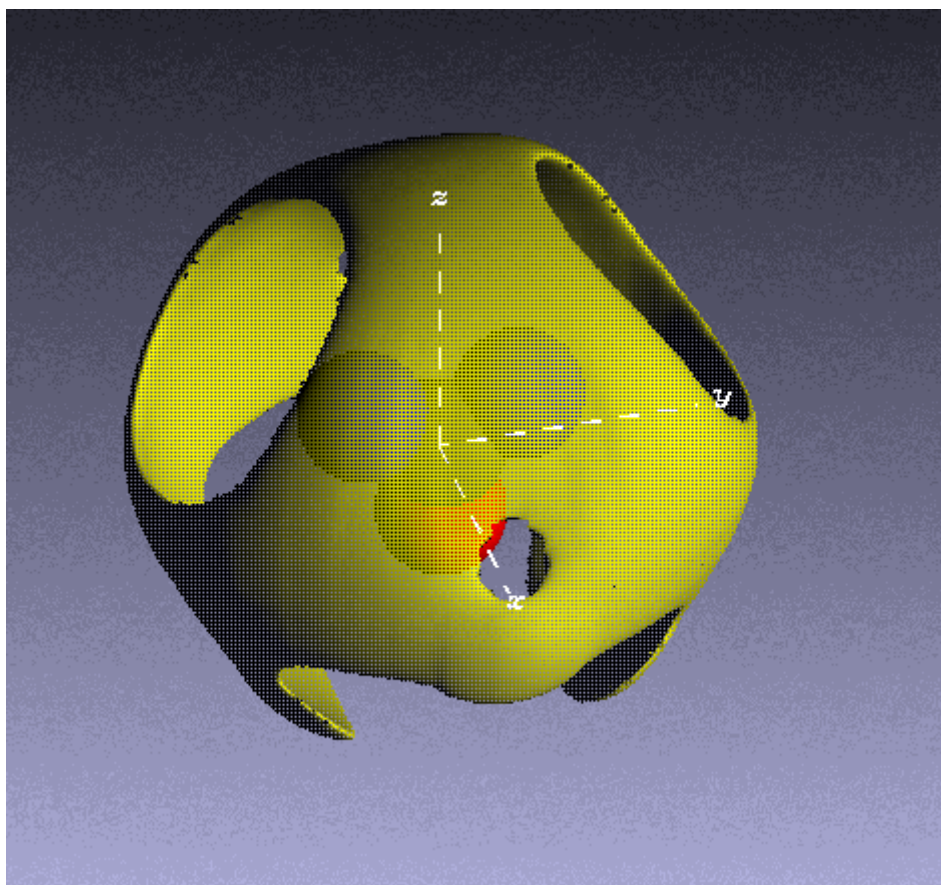


FIGURE S12: Spatial density function of urea (Cu) about urea (Cu) in the TMAO:urea 1:2 solution. The contour level for the isosurface plot includes 20% of the molecules in the distance range $3.5 \text{ \AA} \leq d(\text{Cu-Cu}) \leq 6.0 \text{ \AA}$. The oxygen atom is in red, the two nitrogen atoms in blue.

References

1. Meersman, F., D. Bowron, A.K. Soper and M.H.J. Koch. 2009. Counteraction of urea by trimethylamine N-oxide is due to direct interaction. *Biophys. J.* 97:2559-2566.