Supplementary Information to the manuscript:

Insights into the binding of arginine to adenosine phosphate from mimetic complexes

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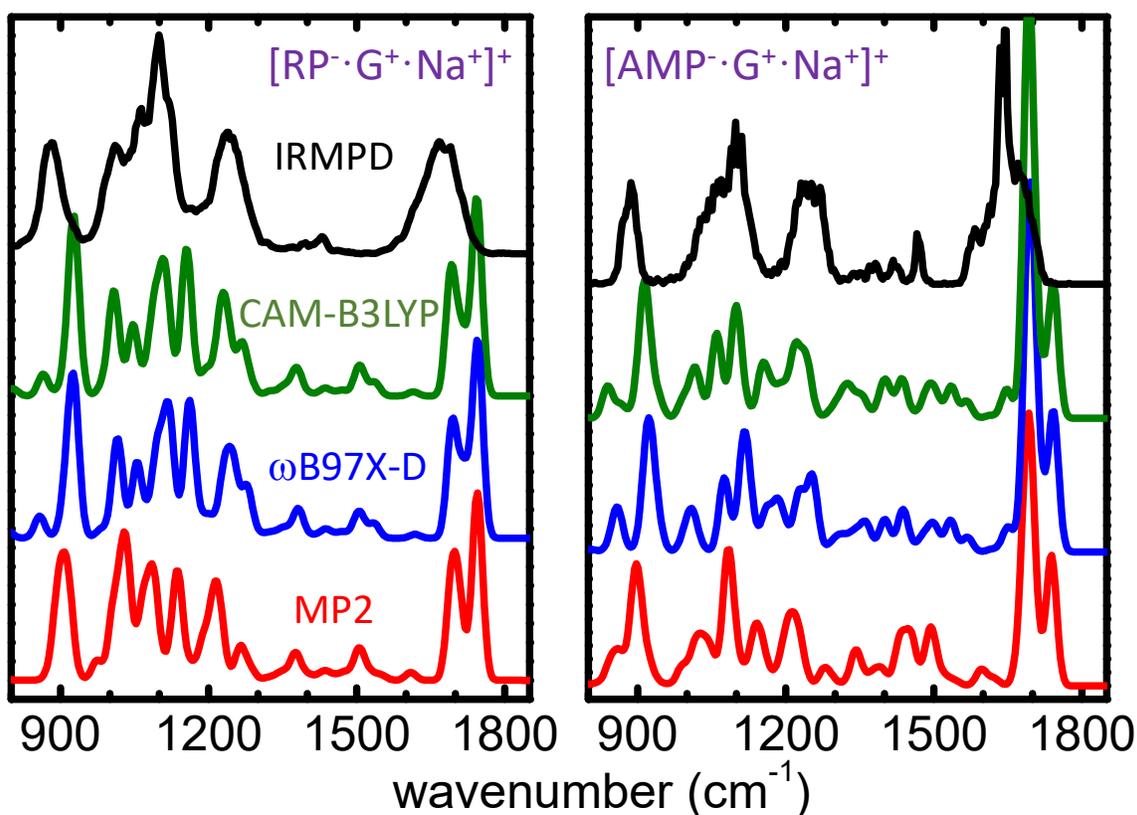## I.   IR SPECTRA PREDICTED BY DIFFERENT COMPUTATIONAL METHODS

FIG. 1.   **Supplementary Information:**   Experimental IRMPD spectra recorded for the $[RP^- \cdot G^+ \cdot Na^+]^+$ and $[AMP^- \cdot G^+ \cdot Na^+]^+$ complexes, and harmonic infrared spectra predicted by the CAM-B3LYP, ωB97X-D and MP2 computations for the low energy conformers R1 and A1, respectively. All computational spectra are displayed without any frequency scaling

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II. REMARKS ON SOLVATION EFFECTS

A recognized advantage of investigating mass-selected complexes in the gas phase is that the supramolecular system under study is precisely defined, so that quantum chemical modelling can be applied to trace back the recorded spectra to host-guest coordination structures. Once this is achieved, predictions must be sought for the link of the observed structures and energetics with the corresponding features in solution. The systems under study here are plausibly strongly interacting with the aqueous solvation shell around the ionic phosphate, guanidinium and sodium ionic moieties. As an approximation to solvation effects, we have computed the net free energies in aqueous solution for the conformers of the RP^- and AMP^- complexes discussed in the previous Sections. Taking into account the relevant stages of the thermodynamic cycle for the solvation process, net free energies in solution may be obtained as $\Delta G_T = \Delta G - \Delta G_{solv}$, where ΔG denotes the conformational free energy in the gas phase and ΔG_{solv} the free energy of solvation [1, 2]. Table I provides the result of the computations performed at the ωB97X-D/6-311++(2d,p) level.

One main finding of these computations is that the conformers in which Na^+ is peripherally bound to the phosphate moiety, $i.e.$, R2 and A3, feature the most favorable solvation energies. This is an interesting feature, as it leads to an enhanced stabilization of the conformations that lie higher in energy in the gas-phase. In fact, the relative free energies predicted for the A1–A3 conformers of the $[AMP^- \cdot G^+ \cdot Na^+]^+$ complex are appreciably altered in solution with respect to the isolated system. The computation yields a solvation free energy for the A3 conformer that is $21\,kJ \cdot mol^{-1}$ greater than those of the A1 and A2 conformers. This can be traced back to the fact that A3 benefits from the more facile access of water solvent molecules to Na^+, whereas the extended 'pocket' built by the phosphate-adenosine backbone in conformers A1 and A2 hides the Na^+ cation, thereby hindering its interaction with the solvent. As a result, the A3 conformer becomes roughly isoenergetic with the A2 one, while A1 remains as the most stable configuration of the complex. A similar trend is observed for the $[RP^- \cdot G^+ \cdot Na^+]^+$ complex. In this case the solvation exothermicity is increased in the R2 conformer vs. the R1 conformer in a more moderate way, by $7\,kJ \cdot mol^{-1}$. The smaller phosphate-ribose backbone (as compared to AMP) produces a smaller 'pocket' region in which the guanidinium and Na^+ cations may still be fairly easily accessed by water molecules, irrespectively of their binding site (on-phosphate or in-pocket, see Fig. 3 of the paper).

The above considerations about the relative stability of the A1 and A3 conformations of the AMP^- complex remain tentative until benchmark experiments are performed in aqueous solution. A hint about what can be expected in solution may be obtained from the inspection of related structures of AMP complexes with arginine residues in the Protein Data Bank (PDB) [3]. We reviewed several tens of complexes of kinases and transferases and found in all cases that the guanidinium side chain binds to the phosphate group of AMP [4]. Nevertheless, the role of the alkali cations present in the PDB cluster structures and of the steric effects induced by the protein architecture remain difficult to assess.

TABLE I. Free energies in aqueous solution, $\Delta G_T = \Delta G - \Delta G_{solv}$ for the conformers explored in this study (in $kJ \cdot mol^{-1}$, relative to the lowest energy conformer), computed at the ωB97X-D/6-311++(2d,p) level within the framework of the thermodynamic cycle described in refs.[1, 2]. Gas phase zero-point and free conformational energies are denoted by ΔE_{zp} and ΔG, respectively; ΔG_{solv} denotes the solvation energy of each conformer.

$[RP^- \cdot G^+ \cdot Na^+]^+$				
conformer	ΔE_{zp}	ΔG	ΔG_{solv}	ΔG_T
R1	0	0	0	0
R2	22	24	-7	17
$[AMP^- \cdot G^+ \cdot Na^+]^+$				
conformer	ΔE_{zp}	ΔG	ΔG_{solv}	ΔG_T
A1	0	0	0	0
A2	8	16	+2	18
A3	37	39	-21	18

[1] T. Dudev, C. Lim, Tetrahedral vs Octahedral Zinc Complexes with Ligands of Biological Interest: A DFT/CDM Study, *J. Am. Chem. Soc.*, 2000, **122**, 11146-11153.

[2] E.F. da Silva, H.F. Svendsen, K.M. Merz, Explicitly Representing the Solvation Shell in Continuum Solvent Calculations, *J. Phys. Chem. A*, 2009, **113**, 6404-6409.

[3] Research Collaboratory for Structural Bioinformatics Protein Data Bank (RCSB-PDB), https://www.rcsb.org/, accessed september 2022.

[4] For examples of protein-AMP complex structures in the PDB data bank, see *e.g.* doi numbers 10.2210/pdb2RIF/pdb (CBS domain protein PAE2072), 10.2210/pdb2ECK/pdb (2ECK Phosphotransferase) or 10.2210/pdb4EAI/pdb (AMPK core transferase).