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Supporting Information

Similarities and Differences between Crystal and Enzyme Environmental Effects on the Electron Density of Drug Molecules

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1. Diffraction experiment, radiation damage

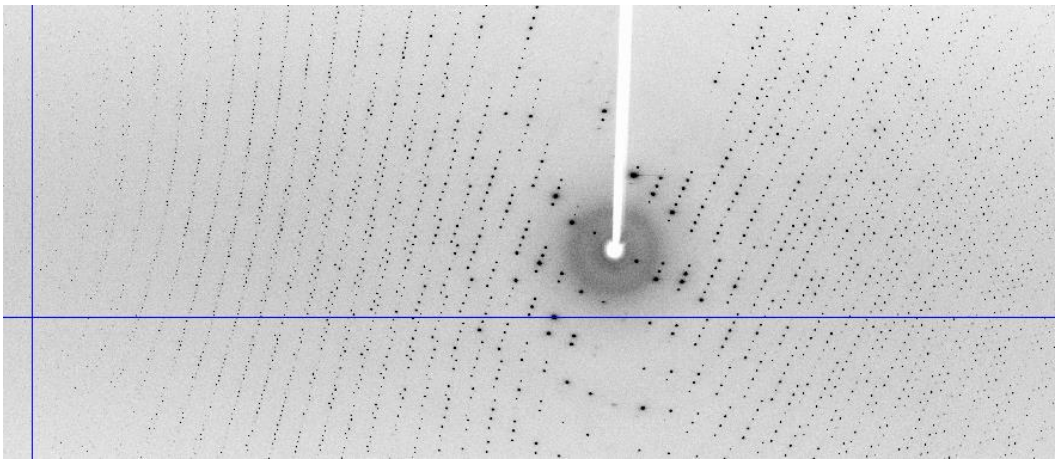

Figure S1: First detection image of **1**K·H_2O measured at BL02B1 of SPring-8 with a curved imaging plate at 25 K. The crosshair of the blue lines indicates a resolution of d=0.220 Å.

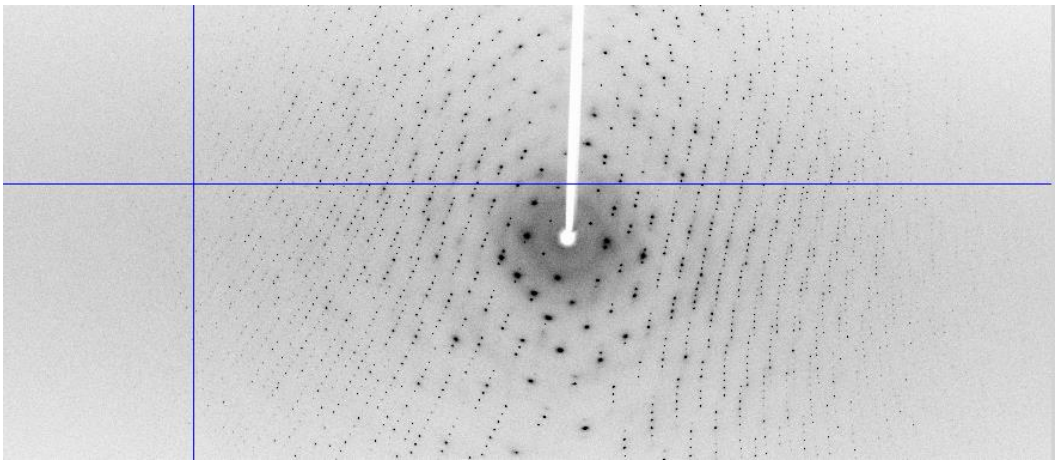

Figure S2: Last detection image of **1**K·H_2O. The crosshair of the blue lines indicates a resolution of d=0.436 Å.

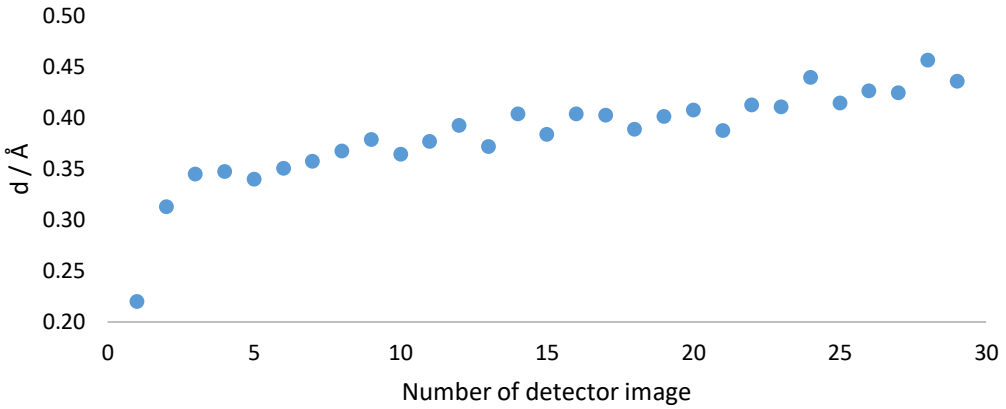

Figure S3: Maximum resolution for the most highly resolved reflection that exceeds the intensity of 190 counts on the imaging plate as a visualization of the radiation damage process.

Table S1: Intensity statistics after correction of radiation damage. The final data set was cut at a maximum resolution of d = 0.45 Å.

Resolution shell (Å)	**No. of measured data**	**Completeness** (%)	**Redundancy**	$\mathbf{R_{merge}}$
Inf-1.44	275	100	7,67	0.0207
1.44-0.95	634	100	7,99	0.0267
0.95-0.75	937	100	7,95	0.0309
0.75-0.66	863	100	7,69	0.0381
0.66-0.60	881	100	7,7	0.0440
0.60-0.55	1060	100	7,55	0.0512
0.55-0.52	863	100	7,31	0.0613
0.52-0.49	1070	100	7,13	0.0713
0.49-0.47	868	100	7,12	0.0869
0.47-0.45	992	100	6,63	0.0971
0.45-0.44	624	100	6,54	0.1106
0.44-0.42	1362	100	6,26	0.1308
0.42-0.41	768	100	6,07	0.1529
0.41-0.40	849	100	5,72	0.1679
0.40-0.39	938	99,9	5,72	0.2013
0.39-0.38	1068	99,9	5,62	0.2239
0.38-0.37	1127	99,7	5,29	0.2539
0.37-0.36	1330	99,1	5,25	0.2912
0.36-0.35	1436	99,2	4,95	0.3472

2. Multipole modeling

Multipole modeling was carried out in the software XD2006 (A Computer Program Package for Multipole Refinement, Topological Analysis of Charge Densities and Evaluation of Intermolecular Energies from Experimental and Theoretical Structure Factors. A. Volkov, P. Macchi, L. J. Farrugia, C. Gatti, P. Mallinson, T. Richter, T. Koritsanszky, 2006). Local coordinate systems were applied as follows (for the atom numbering see Figure 1 in the main manuscript): C1 and C2 without symmetry constraints (*1*); C3, C4, O2, O3, O4, O5, N1 with mirror symmetry (*m*); O1 with *mm2* symmetry; all H atoms with 6-fold symmetry; K1 and K2 with 2-fold symmetry and further constraints due to their special positions (Table S2). O2 was constrained to O3, H11 to H12. Group charges were defined and used as additional constraints, disallowing charge transfer: asymmetric unit (neutral), anion **1** (-1), water (neutral), each K cation (+0.5). κ-parameters were refined individually for all non-H atoms except for O2 that was constrained to O3 and except for both K-atoms, for which κ was kept at 1.0. κ'-parameters were kept at 1.0 for all non-H atoms. For H-atoms, κ/κ' were chosen as 1.3 and 2.0.

Table S2: Chemical constraints for K atoms due to their special positions

	K1	**K2**
Coordinates kept constant	x, z	z
Further positional constraints	none	x = y
Constraints for ADPs	0,5 U11 = U12 0,5 U13 = U23	U11 = U22 U13 = - U23

During the refinement procedure, the scale factor was refined first, followed by monopoles, dipoles and quadrupoles for all atoms except the spherical K atoms. Subsequently, C-H and N-H bond distances in **1** were elongated to values from the QM/MM geometry optimization in the crystal environment (model **C**) in this study (see main text for details). O-H bond distances in water were elongated to values from neutron diffraction (L. A. Malaspina, A. J. Edwards, M. Woinska, D. Jayatilaka, M. J. Turner, J. R. Price, R. Herbst-Irmer, K. Sugimoto, E. Nishibori, S. Grabowsky, *Cryst. Growth Des.* **2017**, *17*, 3812-3825). The H-atom positions were fixed in subsequent refinements, only their isotropic displacement parameters were further refined. For all C-, O- and N-atoms, octupoles and hexadecapoles as well as anisotropic displacement parameters were refined in the final refinement cycles. Final figures of merit of the multipole refinement are: R(F)= 0.0246, $R(F^2)$=0.0393, GooF=3.229.

3. Comparison between theoretical and experimental multipole model and XWR

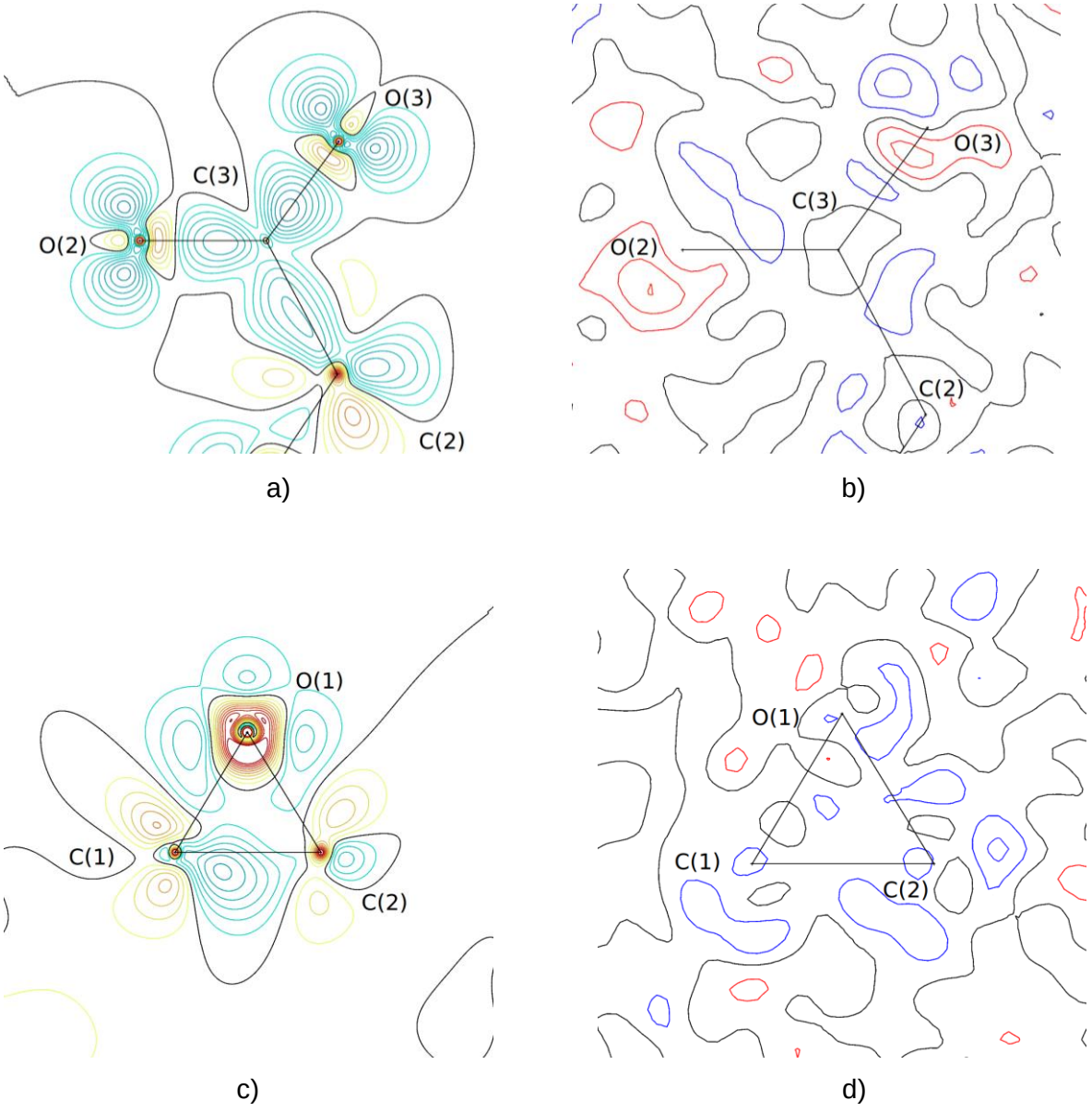

Figure S4: a),c) Static deformation density maps; b),d) residual electron density maps. a),b) Carboxylate group; c),d) epoxide ring. Based on the multipole refinement against the experimental structure factors as described in Section 2. Blue = positive, red = negative. Black = zero contour line. Contour interval = 0.1 eÅ^{-3}.

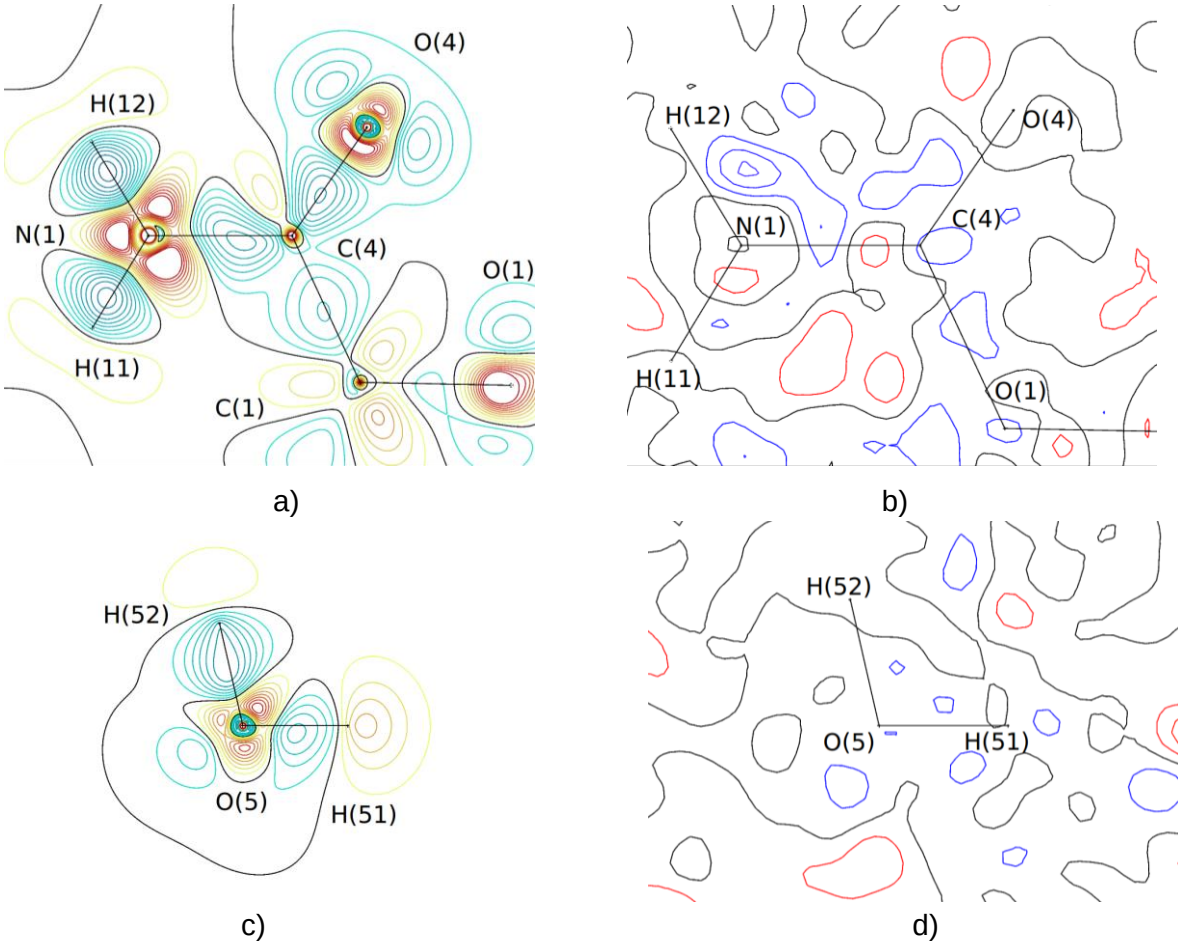

Figure S5: a),c) Static deformation density maps; b),d) residual electron density maps. a),b) Amide group; c),d) water molecule. Based on the multipole refinement against the experimental structure factors as described in Section 2. Blue = positive, red = negative. Black = zero contour line. Contour interval = 0.1 eÅ^{-3}.

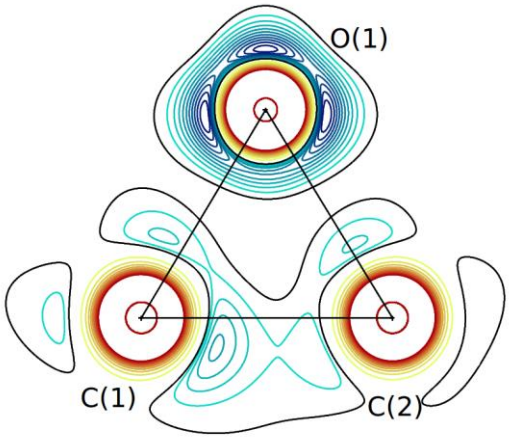

Figure S6: Laplacian map of the epoxide ring. Based on the multipole refinement against the experimental structure factors as described in Section 2. Blue = positive, red = negative. Black = zero contour line. Contour interval = 10 eÅ^{-5} in the positive regions, 100 eÅ^{-5} in the negative regions.

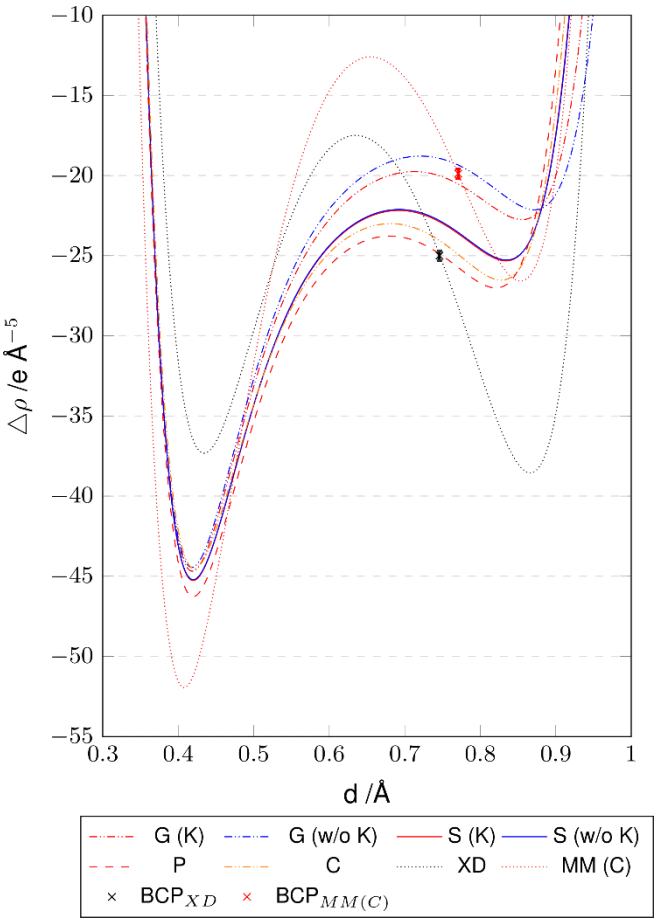

Figure S7. Laplacian of ED along the N1-C4 bond (plotted with the N atom at position 0 Å, left) for the vacuum model (**G**) with or without K^+ counter-cation, the solvent model (**S**) with or without K^+ counter-cation, the crystal model (**C**), the enzyme model (**P**), an experimental (**XD**) and a theoretical (**MM (C)**) multipole model. The theoretical multipole model is based on synthetic structure factors calculated in a pseudo-periodic environment with the software *Denprop*. The theoretical multipole model deviates as strongly from the theoretical models as the experimental multipole model. BCP = bond critical point.

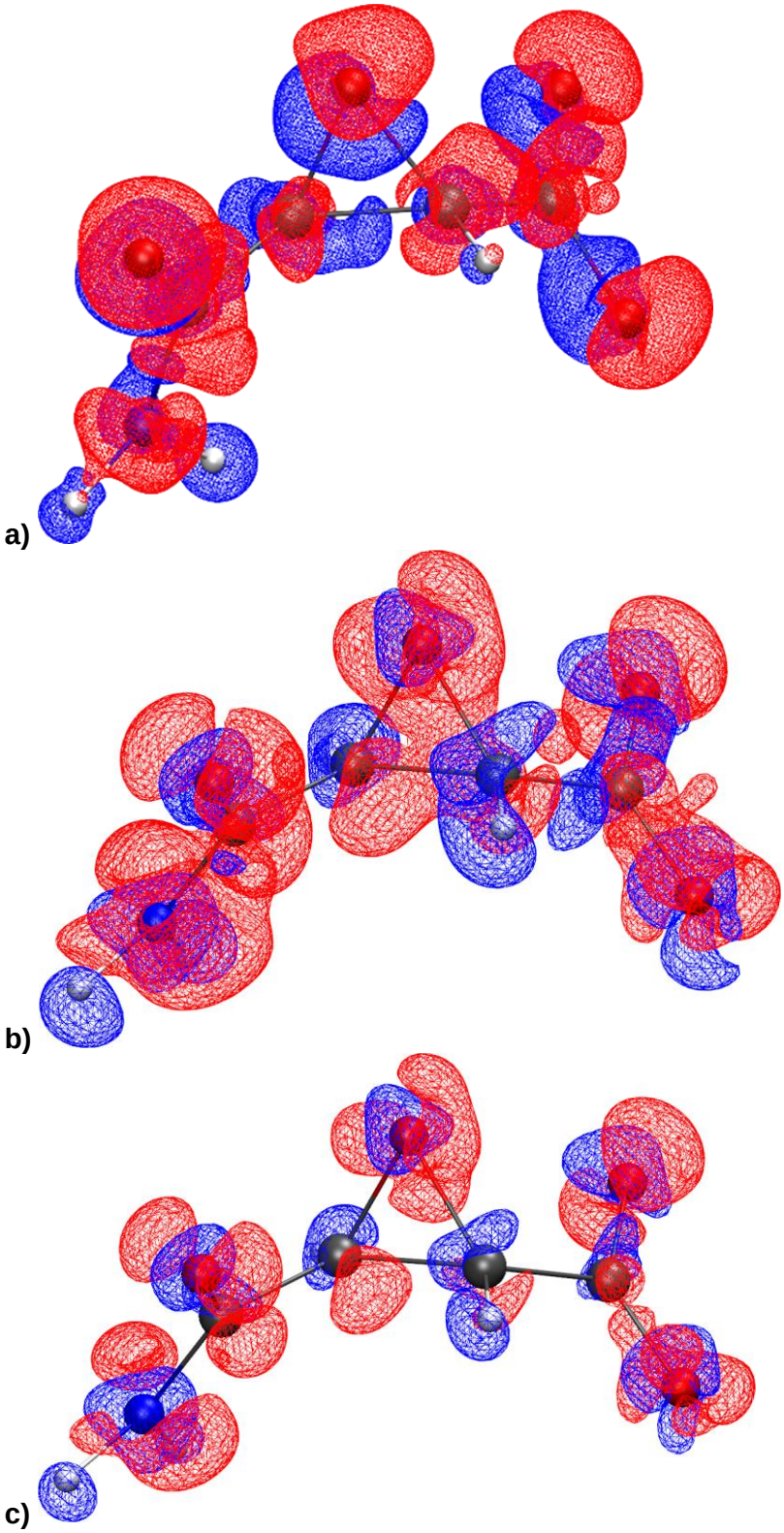

Figure S8. Isosurfaces of the interaction density for a) the experimental multipole model (isovalue ±0.034 *e Å*$^{-3}$), b) the XCW fitting (isovalue ±0.067 *e Å*$^{-3}$), c) the XCW fitting (isovalue ±0.134 *e Å*$^{-3}$). Red regions correspond to higher ED compared to the *in-vacuo* state, blue to lower ED.

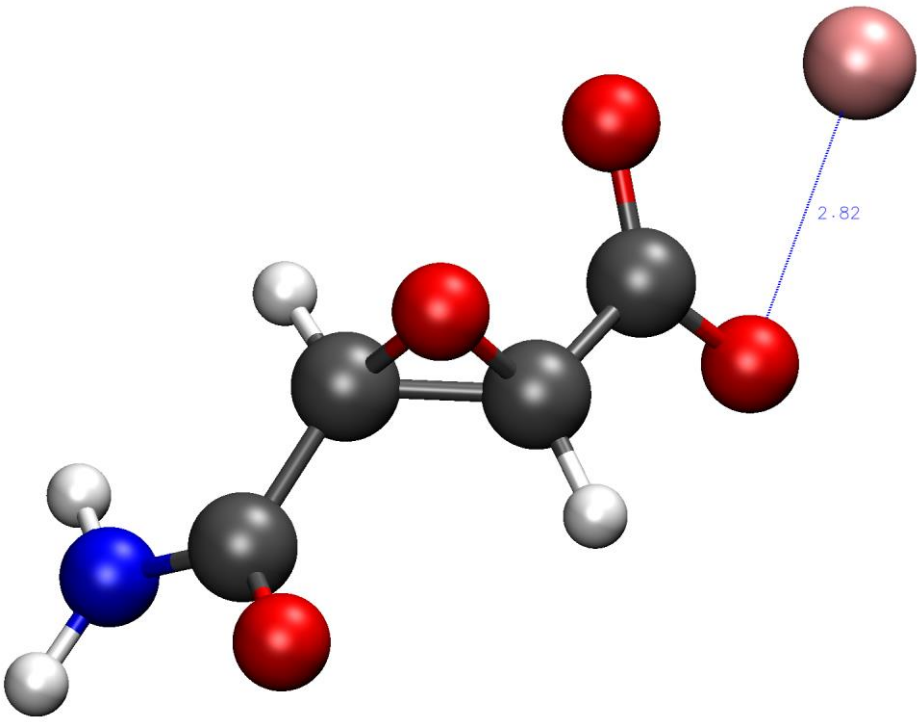

Figure S9: Geometry used in the model **S** (w K). The atomic radii used in the COSMO model are: K 2.223 Å; O 1.720 Å and C 2.000 Å. Therefore, it is ensured the solvation model does not separate the two entities from interacting with each other.

4. Difference in bond densities

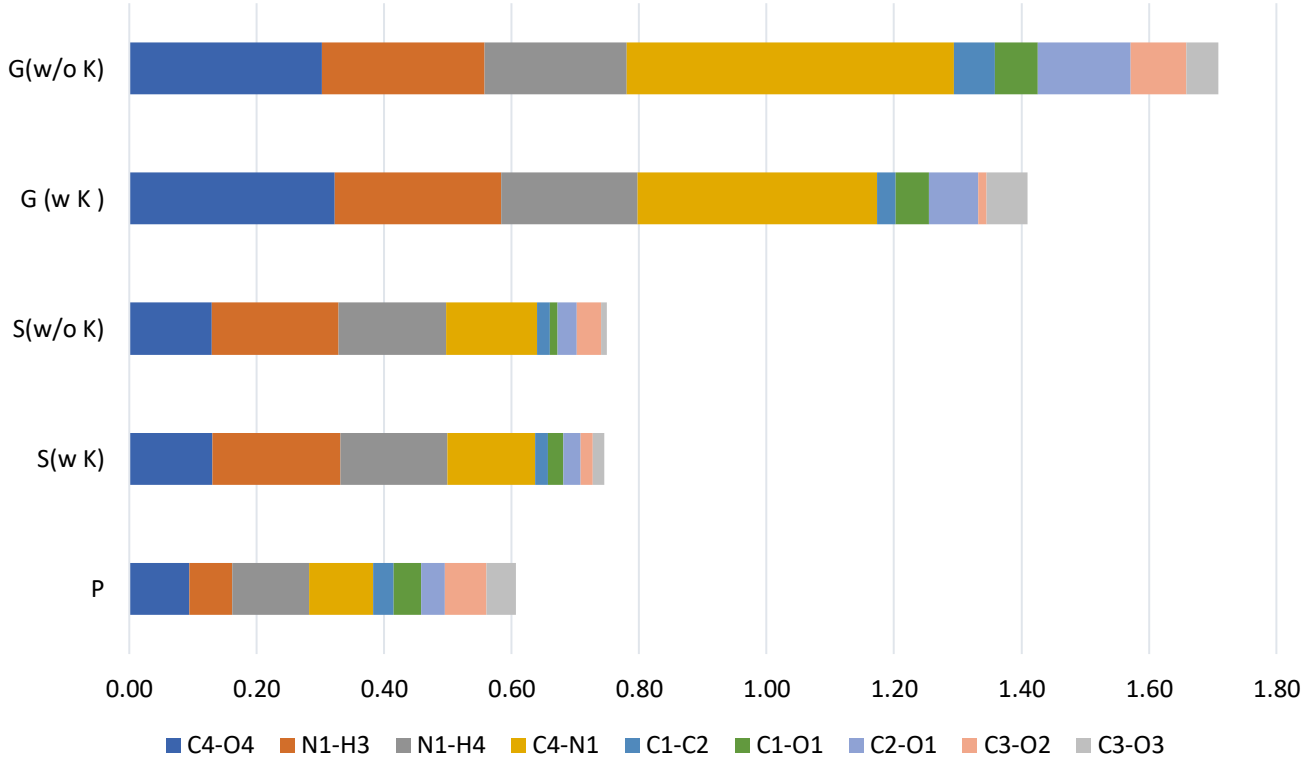

Figure S10: Sum of the number of electrons shifted (N_e in e) for all bond-centered difference density grids. The differences of the vacuum model (**G**) with or without K^+ counter-cation, the solvent model (**S**) with or without K^+ counter-cation and the enzyme model (**P**) are always calculated with respect to the crystal model (**C**).