Supporting information

Perturbation of the conformational dynamics of an active-site loop alters enzyme activity

Donald Gagné[1], Rachel L. French[2], Chitra Narayanan[1], Miljan Simonović[2], Pratul K. Agarwal[3,4] & Nicolas Doucet[1,5,6*]

[1]INRS-Institut Armand-Frappier, Université du Québec, 531 Boul. des Prairies, Laval, Québec H7V 1B7, Canada.

[2]Department of Biochemistry and Molecular Genetics, University of Illinois at Chicago, 900 S. Ashland, Chicago, IL 60607, USA.

[3]Computational Biology Institute and Computer Science and Mathematics Division, Oak Ridge National Laboratory, 1 Bethel Valley Road, Oak Ridge, TN 37830, USA.

[4]Department of Biochemistry, Cellular and Molecular Biology, University of Tennessee, Knoxville, TN 37996, USA.

[5]PROTEO, the Quebec Network for Research on Protein Function, Structure, and Engineering, 1045 Avenue de la Médecine, Université Laval, Québec, QC, G1V 0A6, Canada.

[6]GRASP, the Groupe de Recherche Axé sur la Structure des Protéines, 3649 Promenade Sir William Osler, McGill University, Montréal, QC, H3G 0B1, Canada.

*Contact information: E-mail: nicolas.doucet@iaf.inrs.ca, Phone: (450) 687-5010, ext. 4212.

Correlated motions of residues in the WT and A109G mutant proteins

A comparison of the DCCM difference plots for the reactant state shows enhanced correlated motions of residues 12-18 with 5-11 (C1 in Figure S1A), 41-59 (C2) and 108-119 (C3), with the latter located in the vicinity of the mutation site. These residue groups include the catalytic triad residues H12, K41 and H119, suggesting that the enhanced dynamics of these residues may contribute to the altered catalytic efficiency observed for the mutant. In addition to the correlated motions, several residue groups display anticorrelated motions, such as 35-50 with 58-63 and 73-74; and residues 20-23 with 97-100. Further, lack of correlated motions, observed in the wild-type, were absent between residues forming part of loop 1 (13-17), and loop 4 (63-70), corresponding to A1 in Figure S1A. Conformational dynamics of these loop residues has been shown to be important for catalysis, with truncations of loop 1 and loop 4 residues significantly affecting the catalytic efficiency of RNase A. Our results suggest that correlated motions of these two important loop regions might be important for efficient catalysis.

In contrast to the reactant state, the DCCM difference plots of the product state show more anticorrelations between residues for the mutant relative to the wild-type. These are significant for residues in the N-terminus with other residue groups along the sequence such as 25-43, 56-76 and 91-96. In addition, residues 37-48 showed anticorrelated motions with residues 98-100 (A2 in Figure S1B) and 117-120 (A3). Strong correlated motions were observed between residues 12-14 and residue groups 46-48 (C4 in Figure S1B), 79-82 (C5) and 101-107 (C6) in the mutant relative to the wild-type.

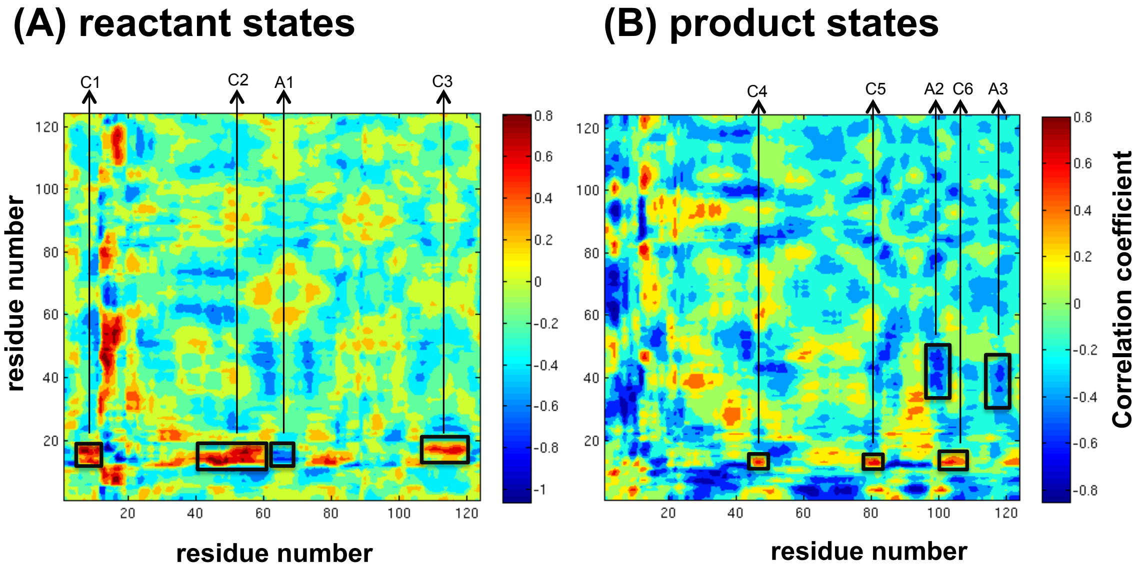

Figure S1 (related to Figure 5). Difference of the dynamic cross-correlation maps (DCCM) between the A09G mutant and wild-type of the bovine RNase A for the reactant (left) and product (right) states. Values closer to 0.8 (red end of the spectrum) represent correlated motions in the mutant relative to the wild-type while values closer to -1 (blue end of the spectrum) represent anticorrelated motions or lack of correlated motions in the mutant that are observed in the wild-type. Regions showing significant correlated and anticorrelated motions are marked as C(1-6) and A(1-3), respectively.