Electronic Supplementary Material (ESI) for Physical Chemistry Chemical Physics.

Supplementary Material:

Computational engineering of cellulase Cel9A-68 functional motions through mutations in its linker region

M. G. S. Costa,[a] Y. F. Silva [a] and P. R. Batista*[a]

[a] *Programa de Computação Científica (PROCC), Fundação Oswaldo Cruz , Rio de Janeiro, Brazil. E-mail: pbatista@fiocruz.br*

Fig. S1.

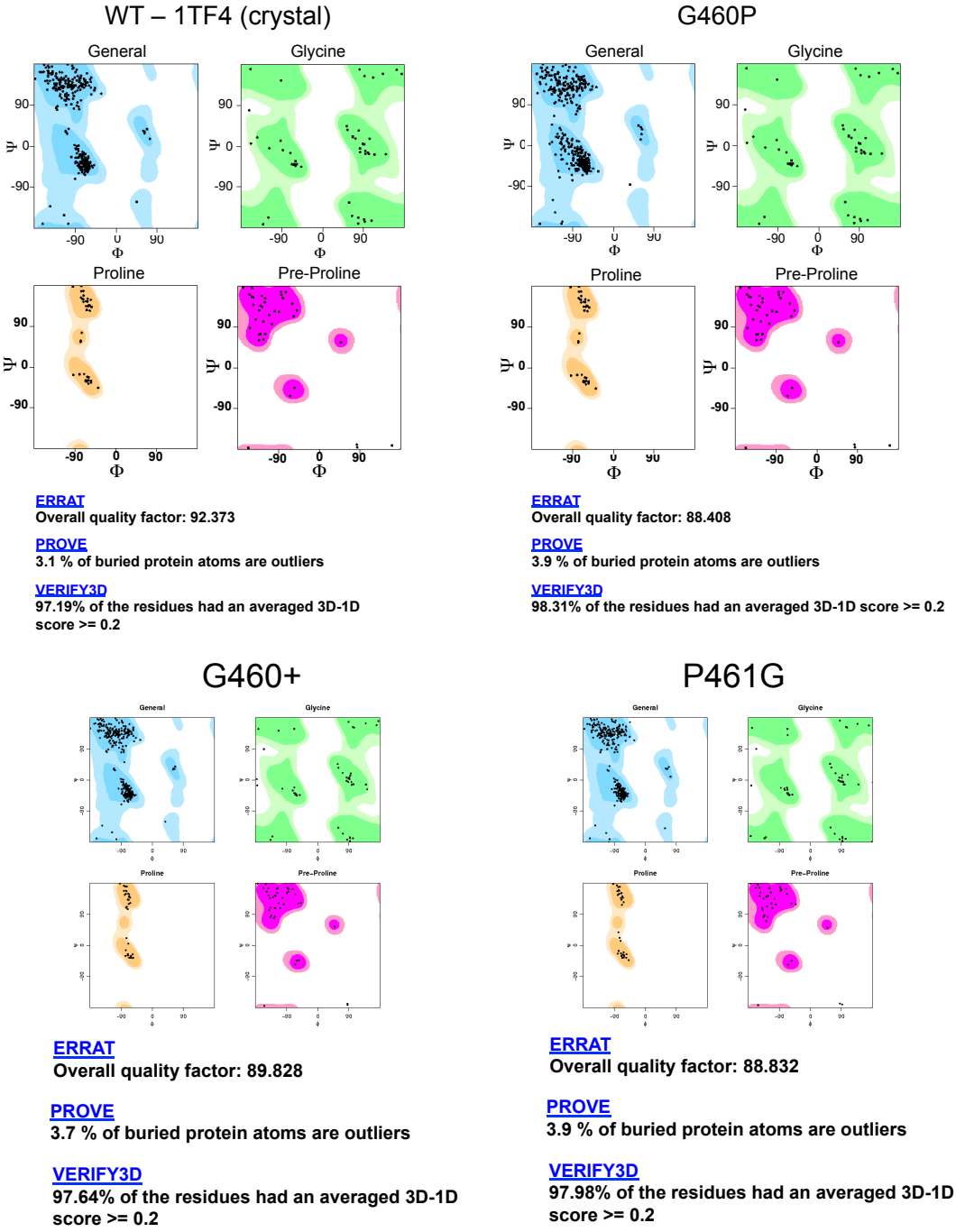

Fig.S1 Validation of the models. The Ramachandran plots and the results returned by ERRAT, PROVE and VERIFY 3D are given for each system considered in this work, including the crystallographic Cel9A-68 structure (PDB ID: 1TF4).

Fig. S2.

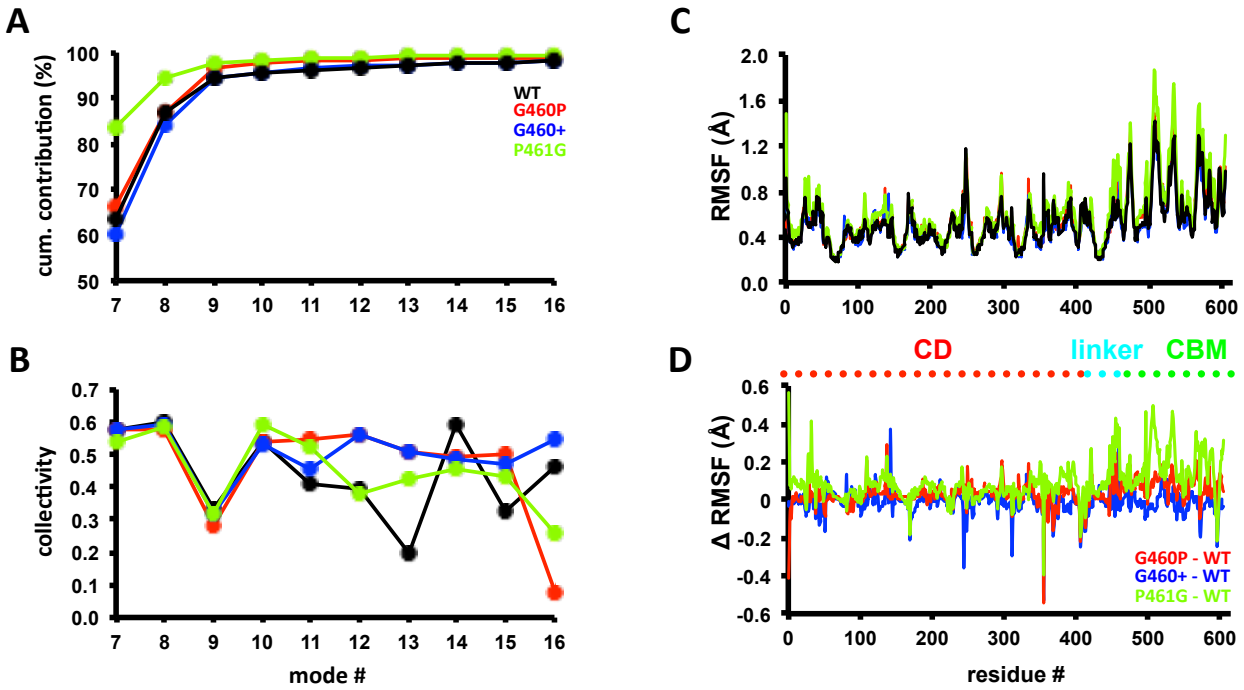

Fig.S2 Normal mode analysis of Cel9A-68 variants. In A, cumulative contribution of the first ten low-frequency normal modes to the overall atomic fluctuations. In B, Collectivity of motions described by low frequency modes. In C, absolute fluctuations per residue. In D, relative fluctuations (taking WT as reference) *per* mode coloured as indicated in the legend. The Cel9A-68 domain definitions are indicated above the plot.

Fig. S3.

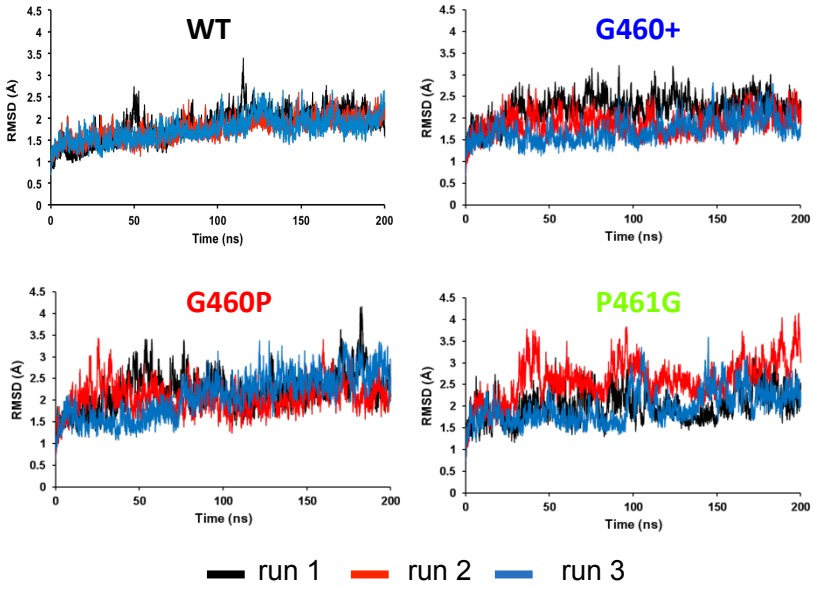

Fig.S3 Time evolution of backbone RMSD *per* replica. The simulated systems are identified above each plot and the results obtained for each independent run are coloured as indicated in the legend.

Fig. S4.

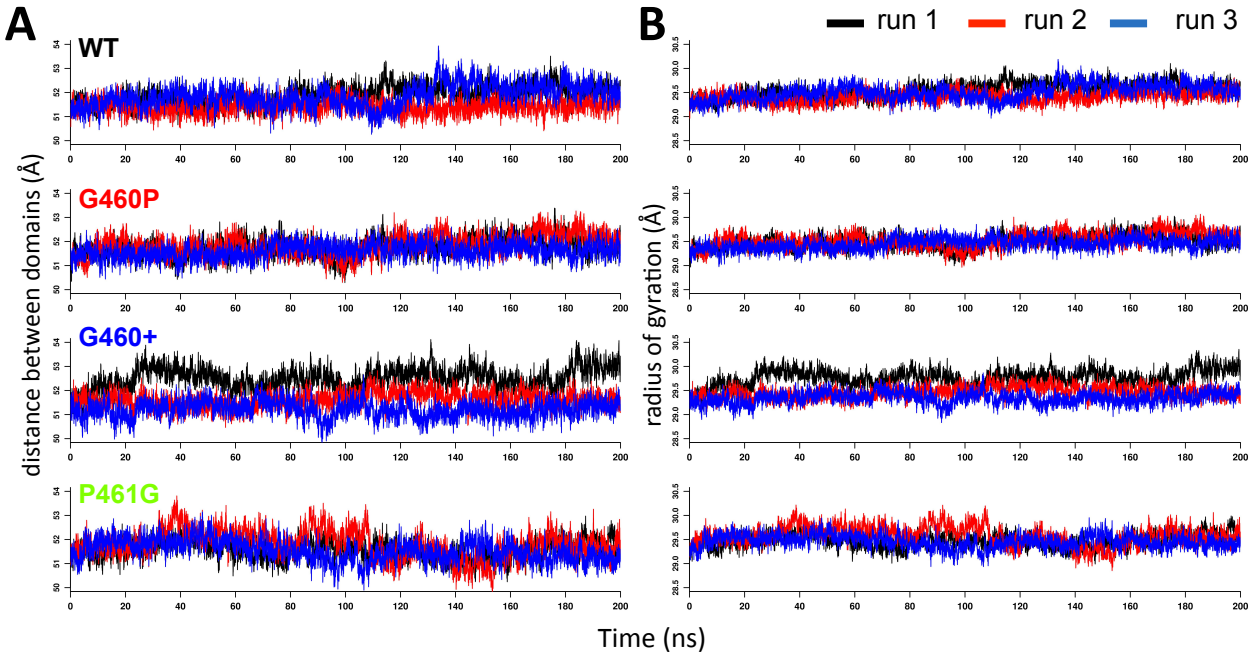

Fig.S4 Time evolution of the interdomain distances (in A) and the radius of gyration of Cel9A-68 (in B), *per* replica, along the MD simulations. The simulated systems are identified above each plot and the results obtained for each independent run are coloured as indicated in the legend.

Fig. S5.

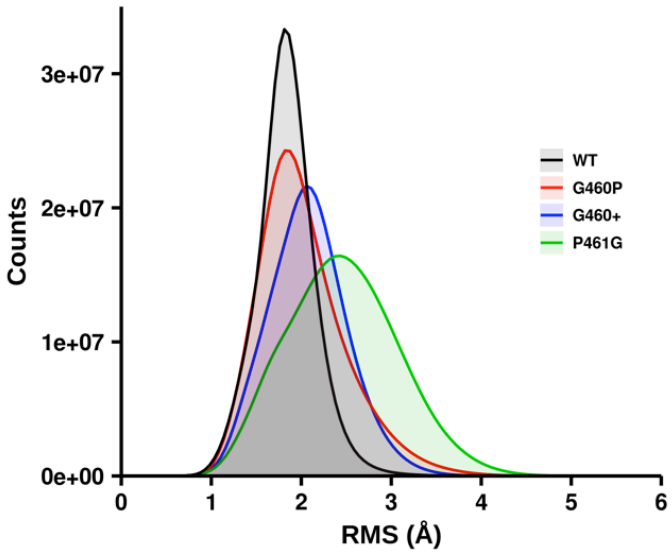

Fig.S5 Distribution of RMSD distances for all pairs of conformations obtained in the concatenated trajectories for each simulated system. Coloured as indicated in the legend.

Fig. S6.

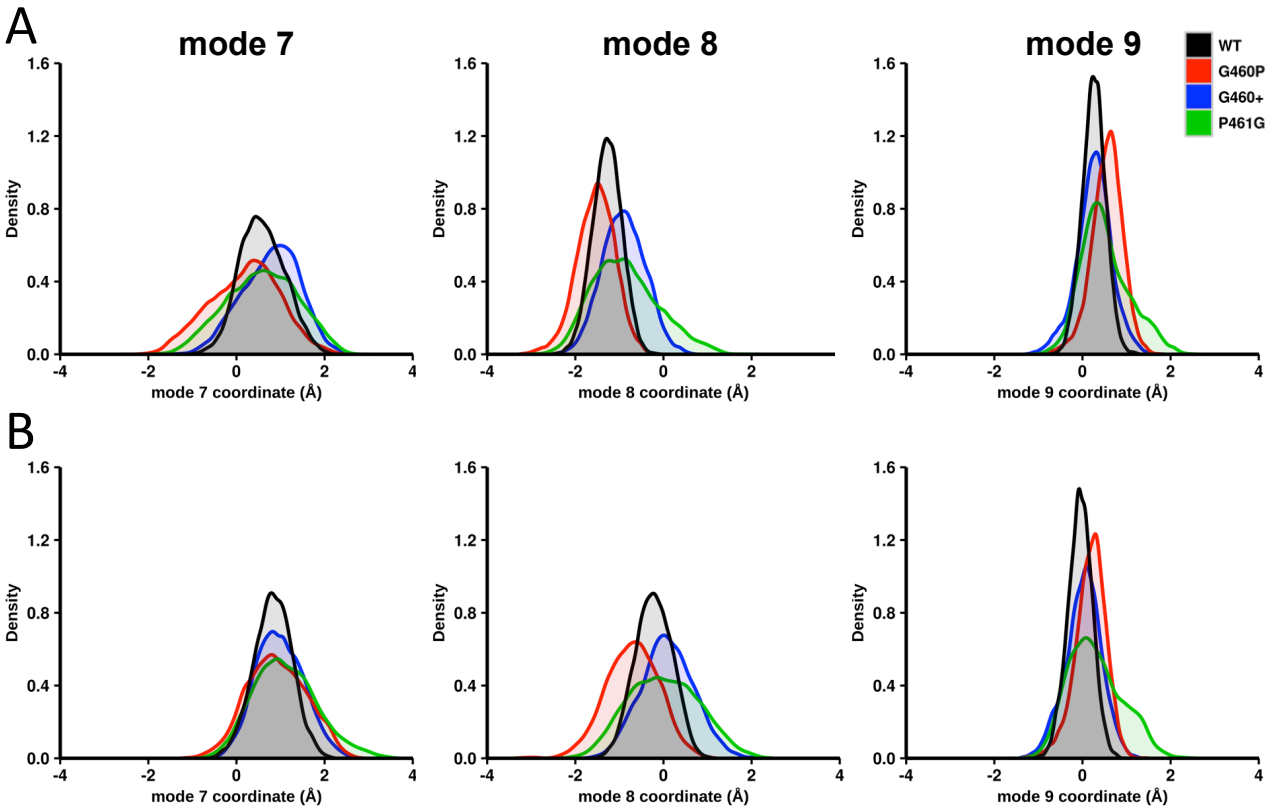

Fig.S6 Results of population analysis based on projections of the conformations sampled during explicit solvent MD onto the three low-frequency normal modes calculated for the WT system (top) or P461G (bottom). Coloured as indicated in the legend.

Captions of supplementary movies

Supplementary movie 1: Displacements along normal mode 7 calculated for the Cel9A-68 wild type system. Each structural domain was differentially coloured as in Figure 1: catalytic domain (red), linker (cyan) and carbohydrate binding module (green).

Supplementary movie 2: Displacements along normal mode 8 calculated for the Cel9A-68 wild type system. The protein is coloured as in supplementary movie 1.

Supplementary movie 3: Displacements along normal mode 9 calculated for the Cel9A-68 wild type system. The protein is coloured as in supplementary movie 1.

Supplementary movie 4: Differences between the lowest frequency normal mode calculated for the WT and P461G systems (related to Figure 3). The Cel9A-68 structural domains are indicated in the left part of the video. Different colouring of the CBM was applied to highlight the conformational states reached after displacements along each sense of the lowest frequency modes.