SUPPLEMENTAL INFORMATION

Molecular Dynamics Study of Functionally Relevant Interdomain and Active Site Interactions in the Autotransporter Esterase EstA from *Pseudomonas aeruginosa*

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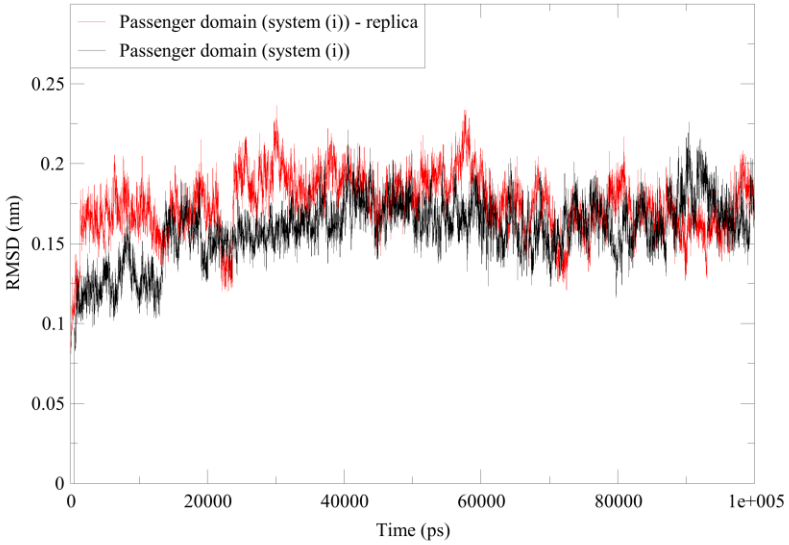
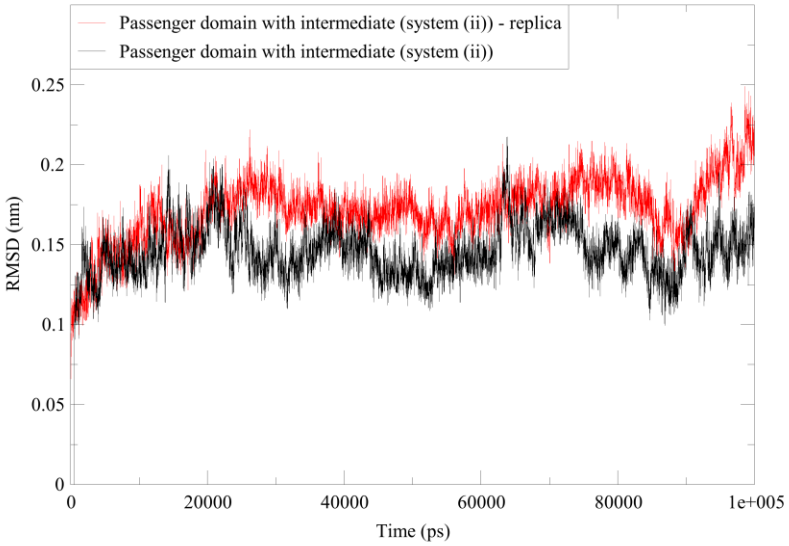
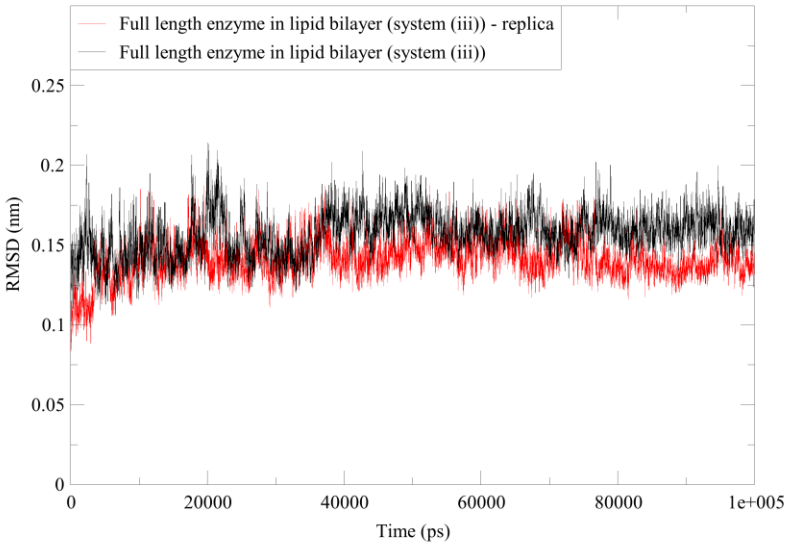
a)



b)


Passenger domain with intermediate (system (ii)) - replica
Passenger domain with intermediate (system (ii))
RMSD (nm)
0.25
0.2
0.15
0.1
0.05
0
0
20000
40000
60000
80000
1e+005
Time (ps)

c)


Full length enzyme in lipid bilayer (system (iii)) - replica
Full length enzyme in lipid bilayer (system (iii))
RMSD (nm)
0.25
0.2
0.15
0.1
0.05
0
0
20000
40000
60000
80000
1e+005
Time (ps)

d)

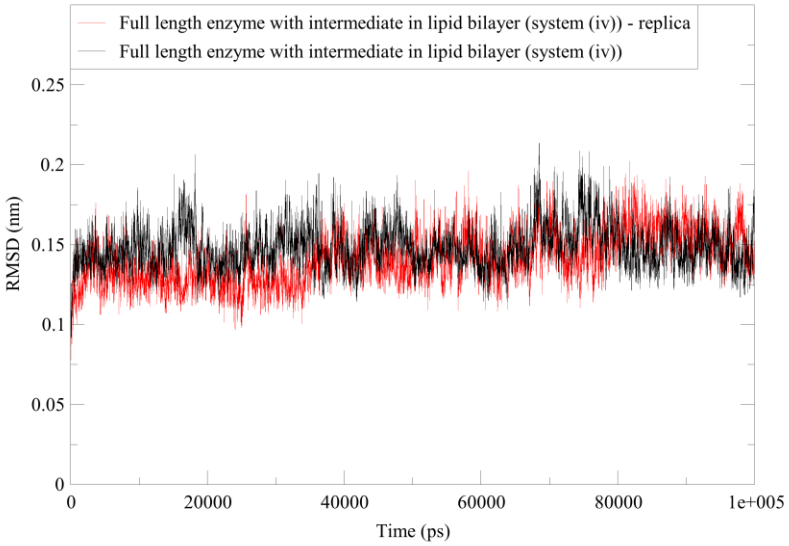
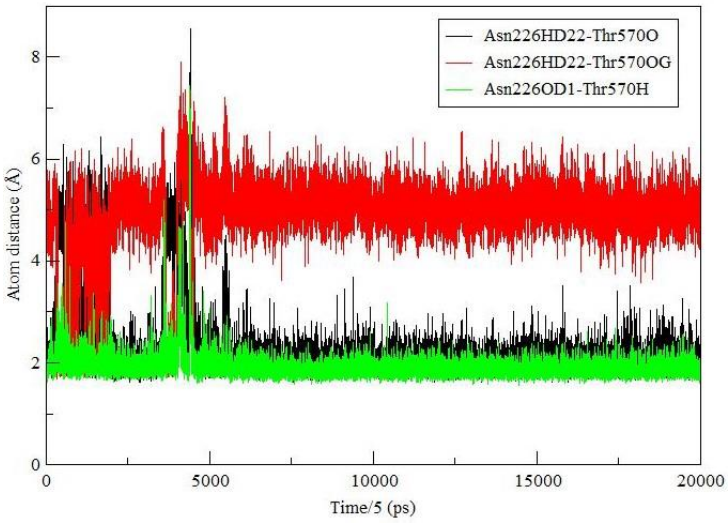
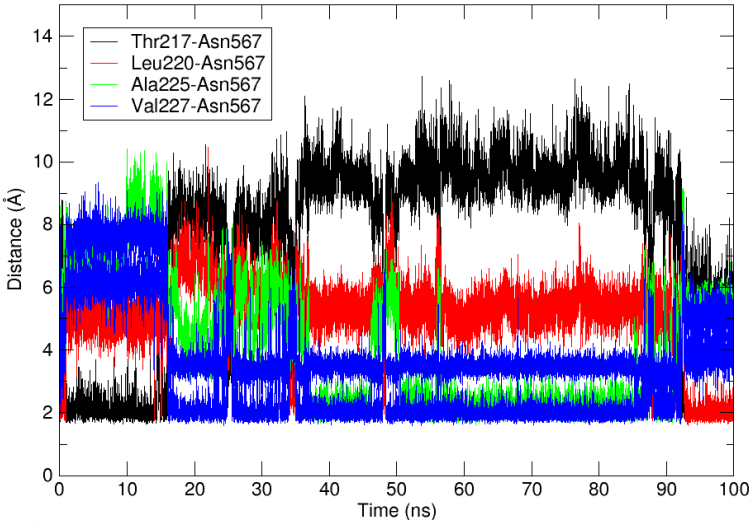
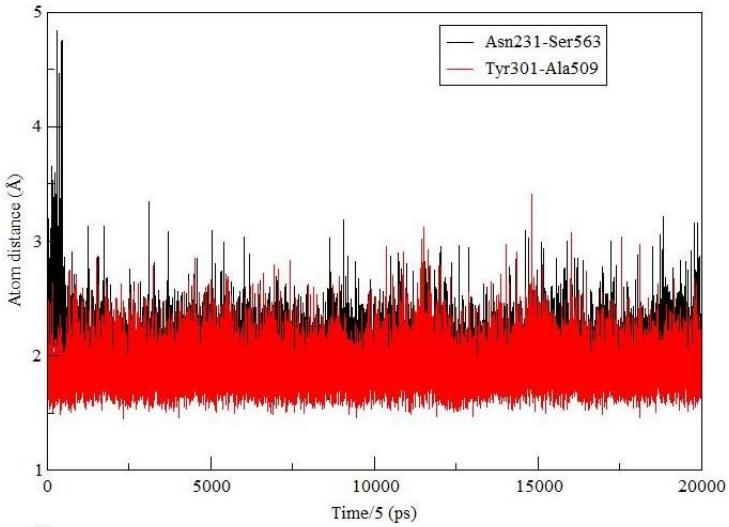


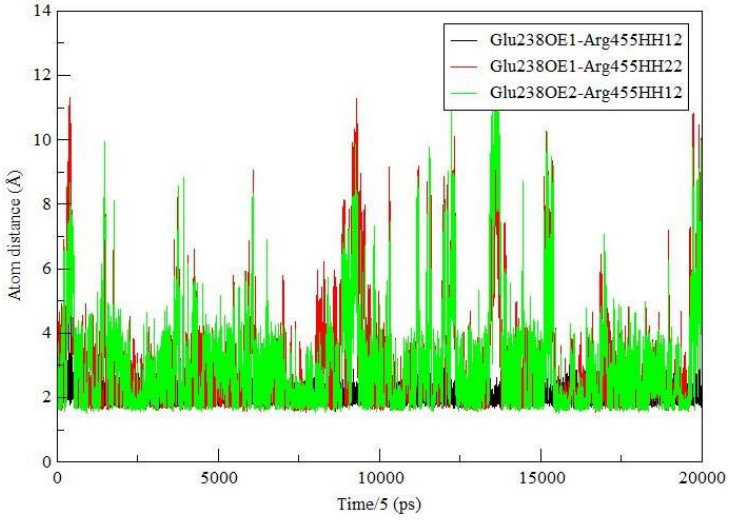
Figure S1. RMSD plots of backbone atoms of the simulations performed twice (black and red) for each of four systems: a) the isolated passenger domain of EstA (system (i)), b) the isolated passenger domain of EstA with 4-hydroxyphenyl octanoate bound as tetrahedral intermediate (system (ii)), c) the full-length EstA enzyme (system (iii)), and d) with (iv) the full-length EstA enzyme with 4-hydroxyphenyl octanoate bound as tetrahedral intermediate (system (iv)).

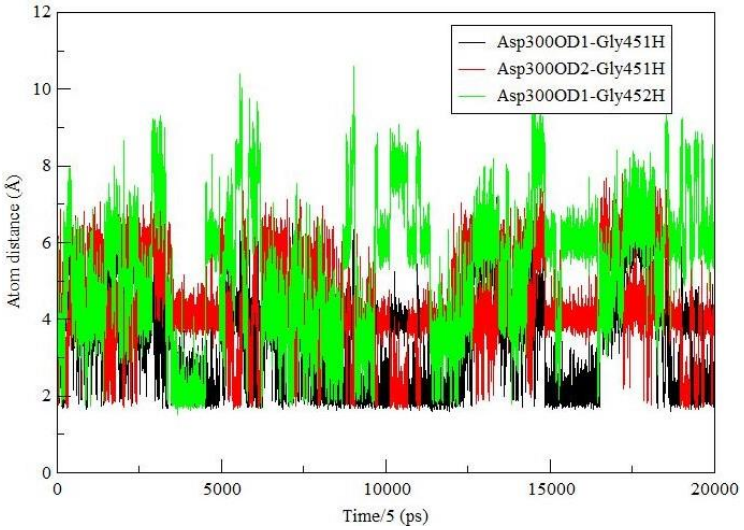
a)



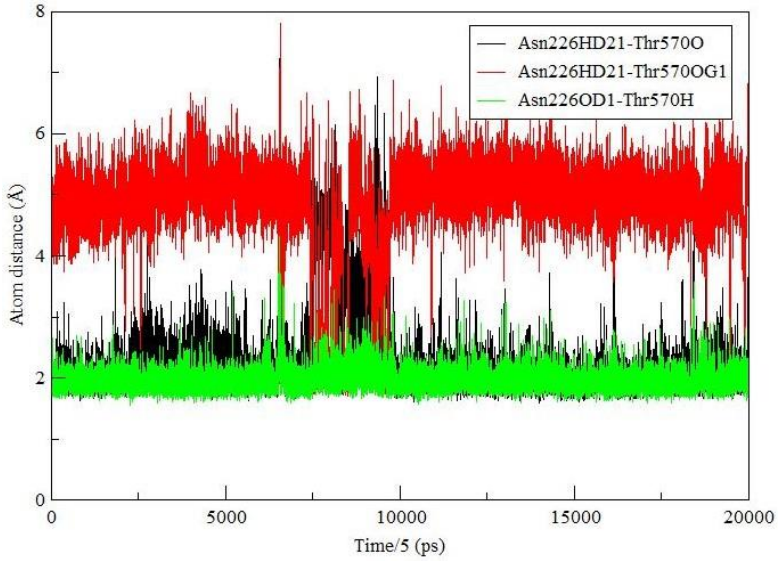

Thr217-Asn567
Leu220-Asn567
Ala225-Asn567
Val227-Asn567
Distance (Å)
Time (ns)

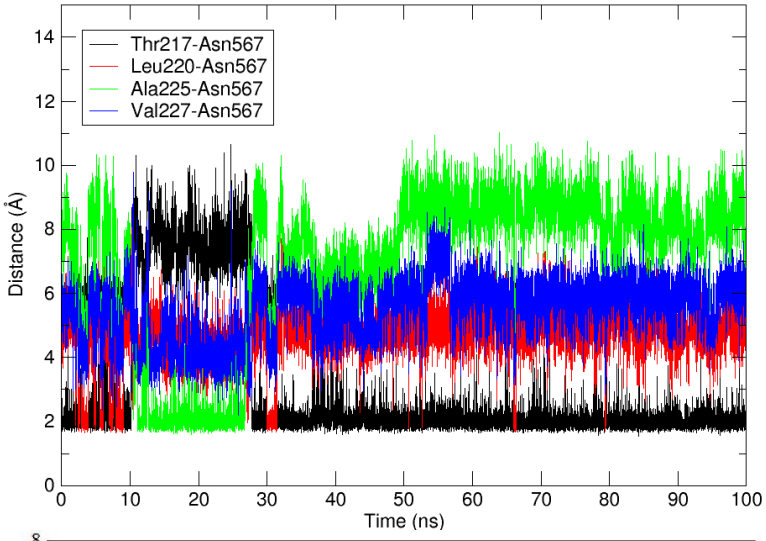

Asn231-Ser563
Tyr301-Ala509
Atom distance (Å)
Time/5 (ps)

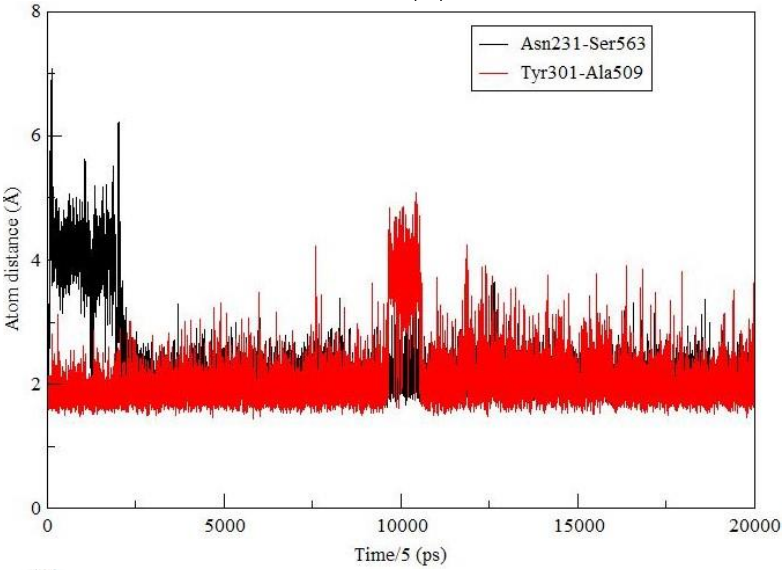

Glu238OE1-Arg455HH12
Glu238OE1-Arg455HH22
Glu238OE2-Arg455HH12
Atom distance (Å)
Time/5 (ps)

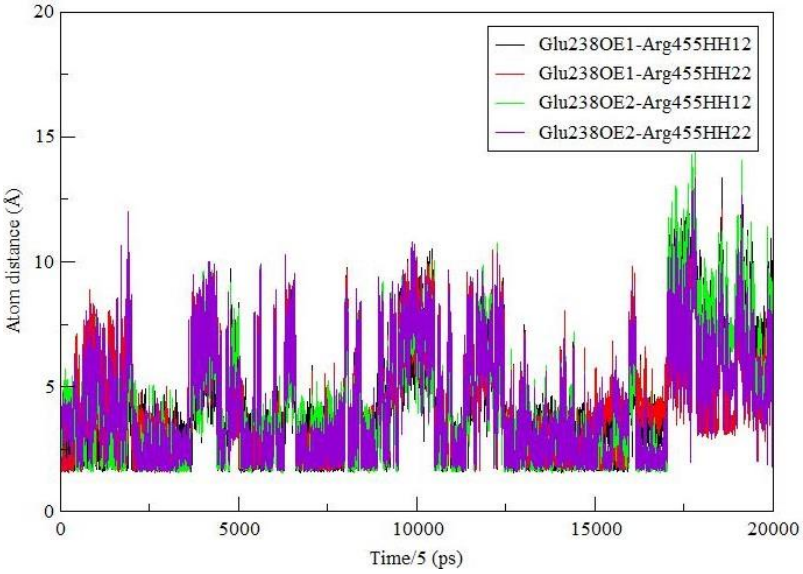

Asp300OD1-Gly451H
Asp300OD2-Gly451H
Asp300OD1-Gly452H
Atom distance (Å)
Time/5 (ps)
0
2
4
6
8
10
12
0
5000
10000
15000
20000

b)


Asn226HD21-Thr570O
Asn226HD21-Thr570OG1
Asn226OD1-Thr570H
Atom distance (Å)
Time/5 (ps)
0
2
4
6
8
0
5000
10000
15000
20000


Thr217-Asn567
Leu220-Asn567
Ala225-Asn567
Val227-Asn567
Distance (Å)
Time (ns)


Asn231-Ser563
Tyr301-Ala509
Atom distance (Å)
Time/5 (ps)


Glu238OE1-Arg455HH12
Glu238OE1-Arg455HH22
Glu238OE2-Arg455HH12
Glu238OE2-Arg455HH22
Atom distance (Å)
Time/5 (ps)

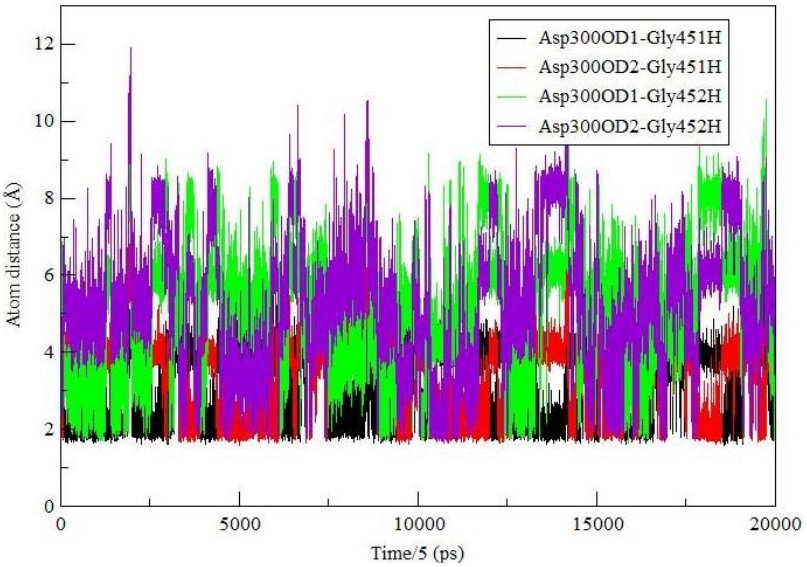


Figure S2. Distance measurements for residues involved in interdomain hydrogen bonds (see Table 1). Atoms involved are specified in the legend only when there are more interacting atoms between two residues. Otherwise, only interacting residues are specified in the legend. a) Full-length enzyme (system (iii)). b) Full-length enzyme with tetrahedral intermediate (system (iv)).

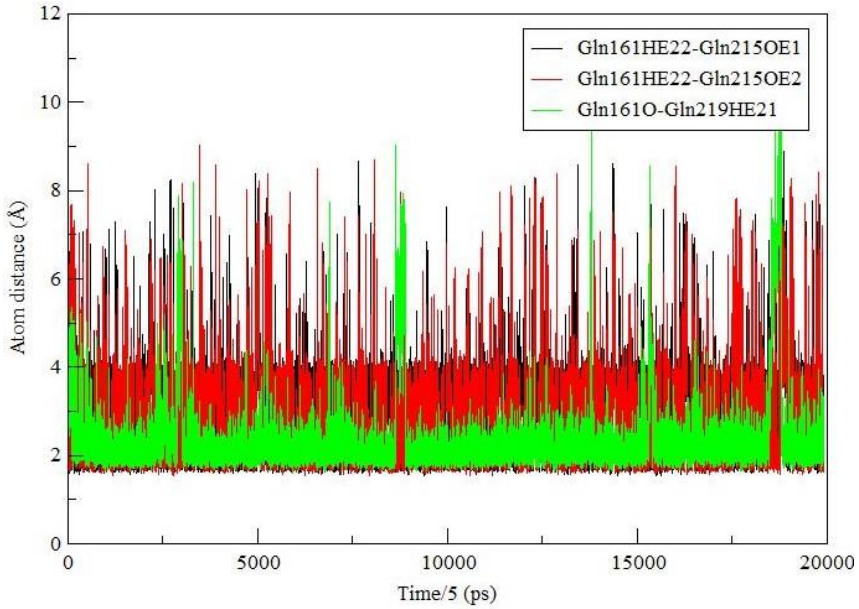


Figure S3. Distance measurement for residues in interaction between helices 7 and 9 in the system of the passenger domain with tetrahedral intermediate (system (ii)).

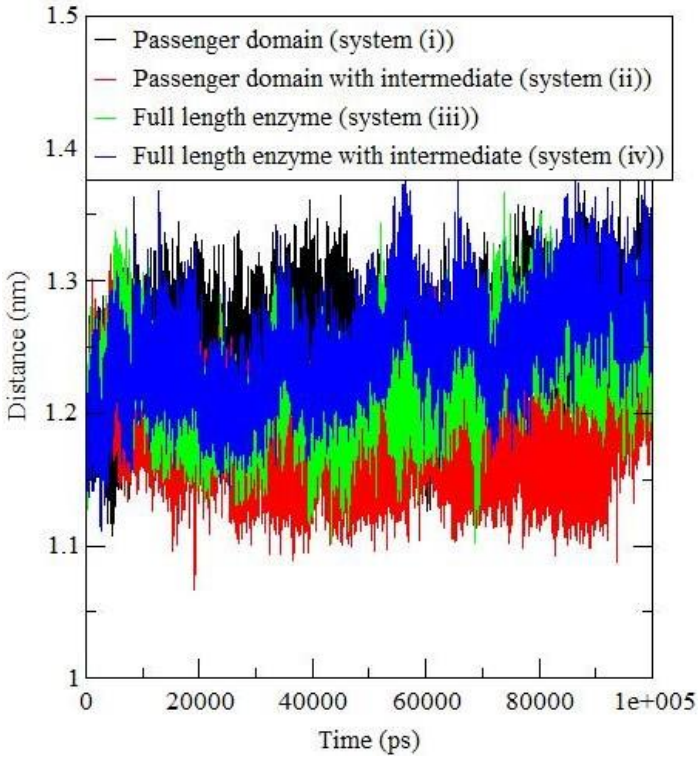
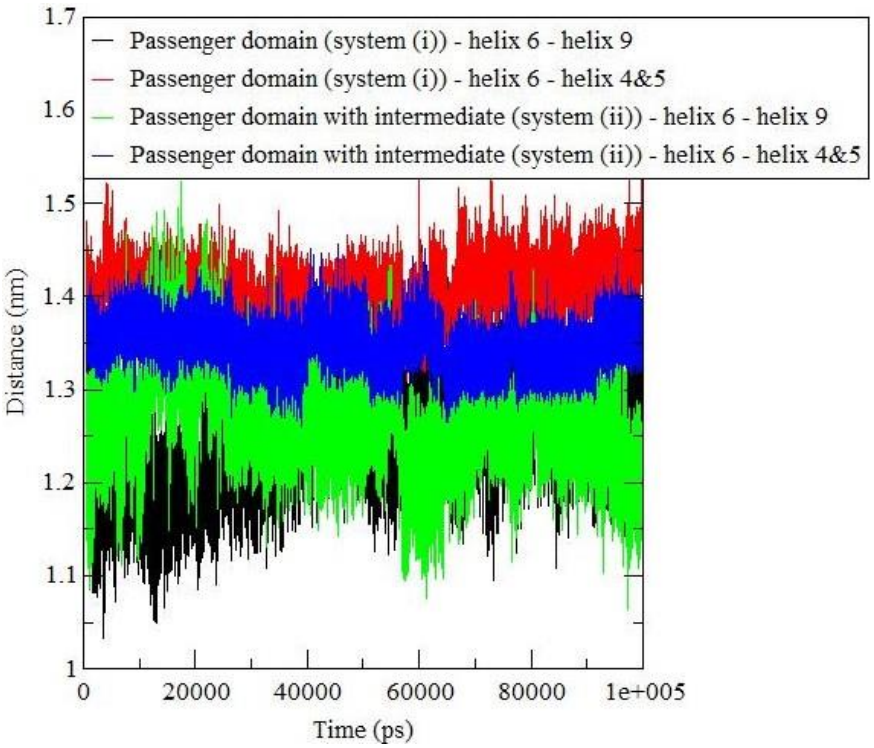


Figure S4. Distance between the centres of mass of helix 6 and the upper part of helix 7 throughout the simulation.



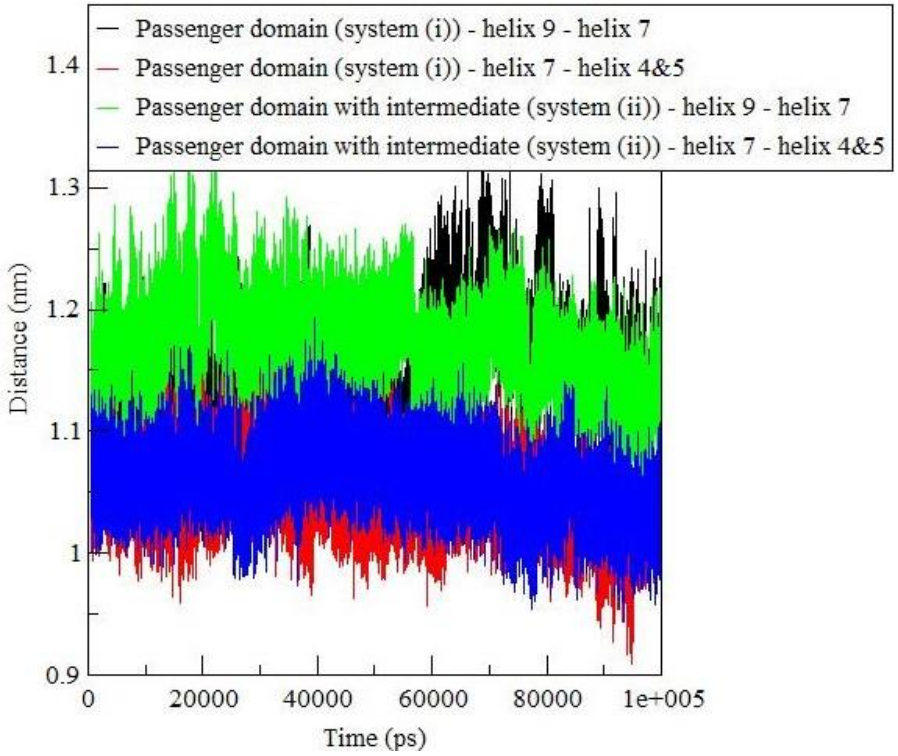


Figure S5. Comparison of centre-of-mass distances between helices in the passenger domain system (i) which exhibits a closed active site conformation and the passenger domain system with bound intermediate (ii) which shows an open active site conformation. a) Distance between helix 6 and helix 9, and distance between helix 6 and helices 4 and 5. b) Distance between helix 9 and helix 7, and distance between helix 7 and helices 4 and 5.

a)

b)

Figure S6. Number of hydrogen bond interactions between water molecules found in the vicinity (3 Å) of DOPE/protein phase and protein. During the simulation of: a) system (iii), b) system (iv).

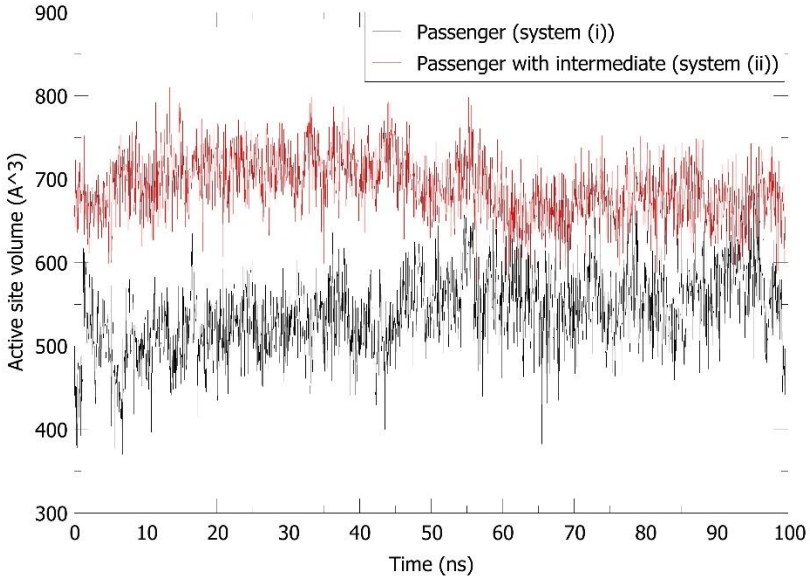


Figure S7. Changes of the active site volume of the isolated passenger domain of EstA (system (i)) (black) and the isolated passenger domain of EstA with intermediate (system (ii))(red). Active site volume calculations were carried out using the POVME 3.0 software [48]. The pocket was defined based on the ligand (4 hydroxyphenyl octanoate) present in system (ii). The inclusion region thus created was used to calculate the pocket volume for system (i) as well.