Supplemental Figures

>FosA^{KP}
MLSGLNHLTLAVSQLAPSVAFYQQLLGMTLHARWDSGAYLSCGDLWLCLSLDPQRRVTPPEES
DYTHYAFSISEADFASFAARLEAAGVAVWKLNRSEGASHYFLDPDGHKLELHVGSLAQRLAAC
REQPYKGMVFFEQ

>FosA^{PA}
MLTGLNHLTLAVADLPASIAFYRDLLGFRLEARWDQGAYLELGSLWLCLSREPQYGGPAADYT
HYAFGIAAADFARFAAQLRAHGVREWKQNRSEGDSFYFLDPDGHRLEAHVGDLRSRLAACRQA
PYAGMRFAD

>FosA3
MLQGLNHLTLAVSDLASSLAFYQQLPGMRLHASWDSGAYLSCGALWLCLSLDEQRRKTPPQES
DYTHYAFSVAEEEFAGVVALLAQAGAEVWKDNRSEGASYYFLDPDGHKLELHVGNLAQRLAA
CRERPYKGMVFFD

>FosA$^{PA}_{c}$
MLTGLNHLTLAVADLPASIAFYRDLLGFRLEARWDQGAYLELGSLWLCLSLDPQRRVTPPEESD
YTHYAFGIAAADFARFAAQLRAHGVREWKQNRSEGDSFYFLDPDGHRLEAHVGDLRSRLAACR
QAPYAGMRFAD

>FosA$^{KP}_{c}$
MLSGLNHLTLAVSQLAPSVAFYQQLLGMTLHARWDSGAYLSCGDLWLCLSREPQYGGPAADY
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PYKGMVFFEQ

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FosA^PA     MLTGLNHLTLAVADLPASIAFYRDLLGFRLEARWDQGAYLELGSLWLCLSREPQYGG---57
FosA^PA_c   MLTGLNHLTLAVADLPASIAFYRDLLGFRLEARWDQGAYLELGSLWLCLSLDPQRRVTPP60
FosA3       MLQGLNHLTLAVSDLASSLAFYQQLPGMRLHASWDSGAYLSCGALWLCLSLDEQRRKTPP60
FosA^KP     MLSGLNHLTLAVSQLAPSVAFYQQLLGMTLHARWDSGAYLSCGDLWLCLSLDPQRRVTPP60
FosA^KP_c   MLSGLNHLTLAVSQLAPSVAFYQQLLGMTLHARWDSGAYLSCGDLWLCLSREPQYGG---57
            ** *********::*  *:***::* *: *.* **.****. * ****** : *

FosA^PA     PAADYTHYAFGIAAADFARFAAQLRAHGVREWKQNRSEGDSFYFLDPDGHRLEAHVGDLR 117
FosA^PA_c   EESDYTHYAFGIAAADFARFAAQLRAHGVREWKQNRSEGDSFYFLDPDGHRLEAHVGDLR 120
FosA3       QESDYTHYAFSVAEEEFAGVVALLAQAGAEVWKDNRSEGASYYFLDPDGHKLELHVGNLA 120
FosA^KP     EESDYTHYAFSISEADFASFAARLEAAGVAVWKLNRSEGASHYFLDPDGHKLELHVGSLA 120
FosA^KP_c   PAADYTHYAFSISEADFASFAARLEAAGVAVWKLNRSEGASHYFLDPDGHKLELHVGSLA 117
              :*******.::  :** ..* *   *.  ** ***** *.********:** ***.*

FosA^PA     SRLAACRQAPYAGMRFAD-    135
FosA^PA_c   SRLAACRQAPYAGMRFAD-    138
FosA3       QRLAACRERPYKGMVFFD-    138
FosA^KP     QRLAACREQPYKGMVFFEQ    139
FosA^KP_c   QRLAACREQPYKGMVFFEQ    136
             .******: ** ** * :
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Figure S1. FosA sequences and alignment of wild-type and chimeric enzymes, with dimer-interface loops highlighted in yellow.

Bonds	**k_b (kcal mol^{-1} Å^{-2})**	**b_0 (Å)**
MN2 - NR2	200.0	2.18
Angles	**k_θ (kcal mol^{-1} rad^{-2})**	**θ_0 (degrees)**
CPH2 - NR2 - MN2	20.0	120.65
CPH1 - NR2 - MN2	20.0	125.98
NR2 - MN2 - NR2	100.0	114.00

Table S1. Force field parameters for Mn^{2+} coordination.

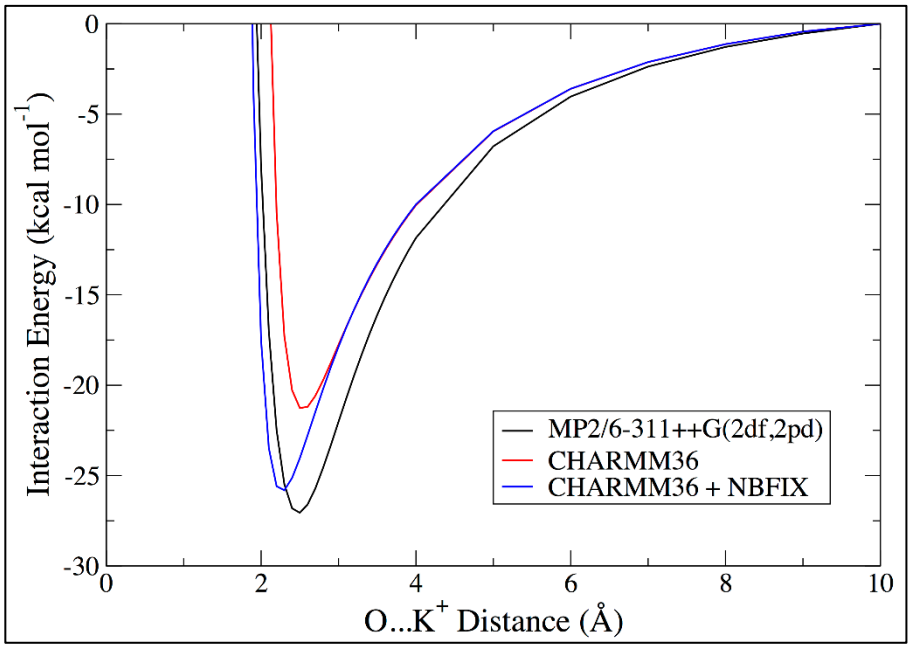

Figure S2. Interaction energy between NMA and a K^+ ion as a function of the distance between the ion and the carbonyl O atom.

	ε (kcal mol^{-1})	**R_{min} (Å)**
POT - O (combination rule)	-0.102176	3.46375
POT - O (NBFIX)	-0.050218	3.30375

Table S2. Combination rule and NBFIX parameters for carbonyl O - K^+ Interaction.

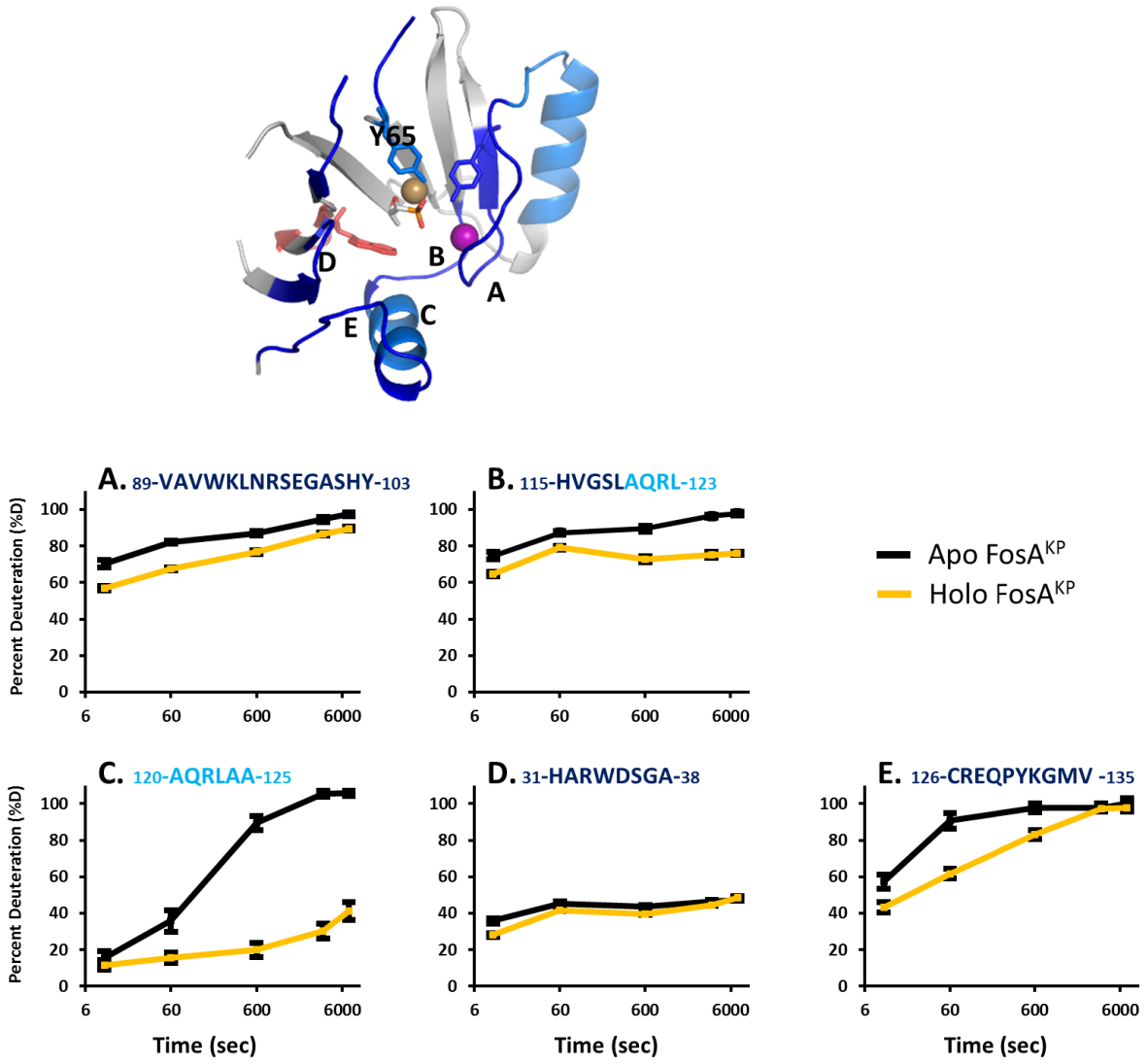

Figure S3. Fosfomycin-induced protection of hydrogen-deuterium exchange near the active site. Peptides displaying significant differences in deuterium uptake are mapped on the crystal structure of $FosA^{KP}$. Regions with statistically significant decreases in deuterium at the earliest deuterium incubation time point (10 sec) are colored dark blue. Regions with decreases in deuterium uptake only observed at later time points are colored light blue. Regions with fosfomycin-induced increase in deuterium uptake are colored red. Important residues are shown as sticks, along with Mn^{2+} (gold), K^{+} (purple), and fosfomycin (sticks). Panel A through E show deuterium uptake traces for representative peptidic fragments in apo and holo $FosA^{KP}$. The sequences of the peptides are shown above each panels.

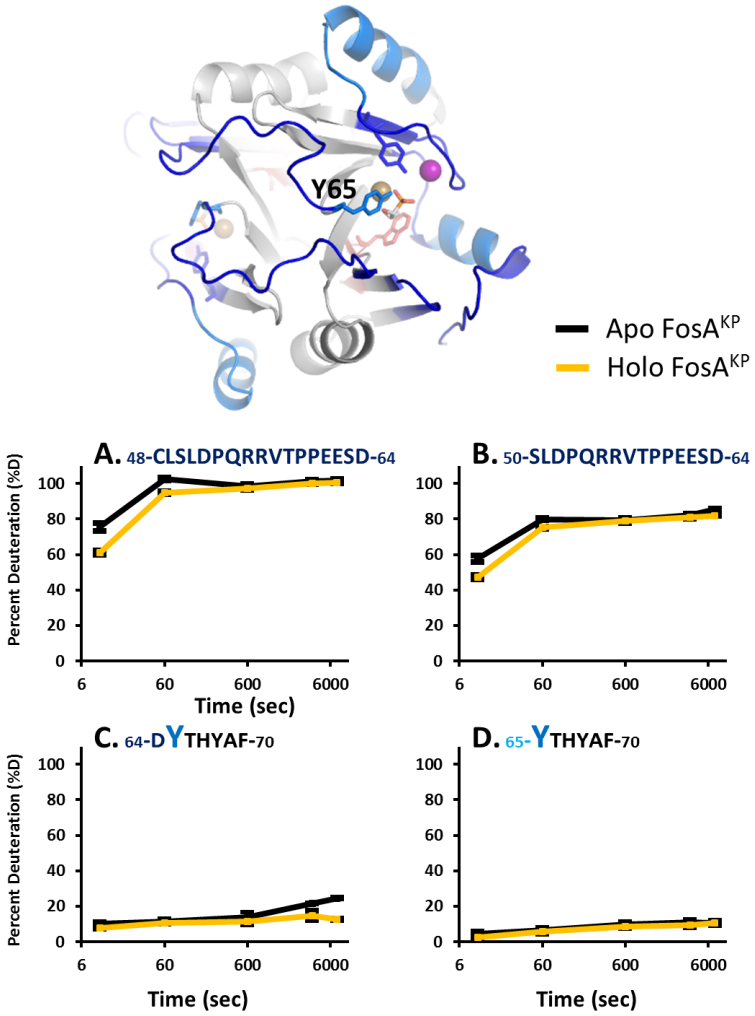

Figure S4. Fosfomycin-induced protection of hydrogen-deuterium exchange at the dimer interface. Peptides displaying significant differences in deuterium uptake are mapped on the crystal structure of $FosA^{KP}$. Regions with statistically significant decreases in deuterium at the earliest deuterium incubation time point (10 sec) are colored dark blue. Regions with decreases in deuterium uptake only observed at later time points are colored light blue. Regions with fosfomycin-induced increase in deuterium uptake are colored red. Important residues are shown as sticks, along with Mn^{2+} (gold), K^{+} (purple), and fosfomycin (sticks). Panel A through D show deuterium uptake traces for representative peptidic fragments in apo and holo $FosA^{KP}$. The sequences of the peptides are shown above each panels.

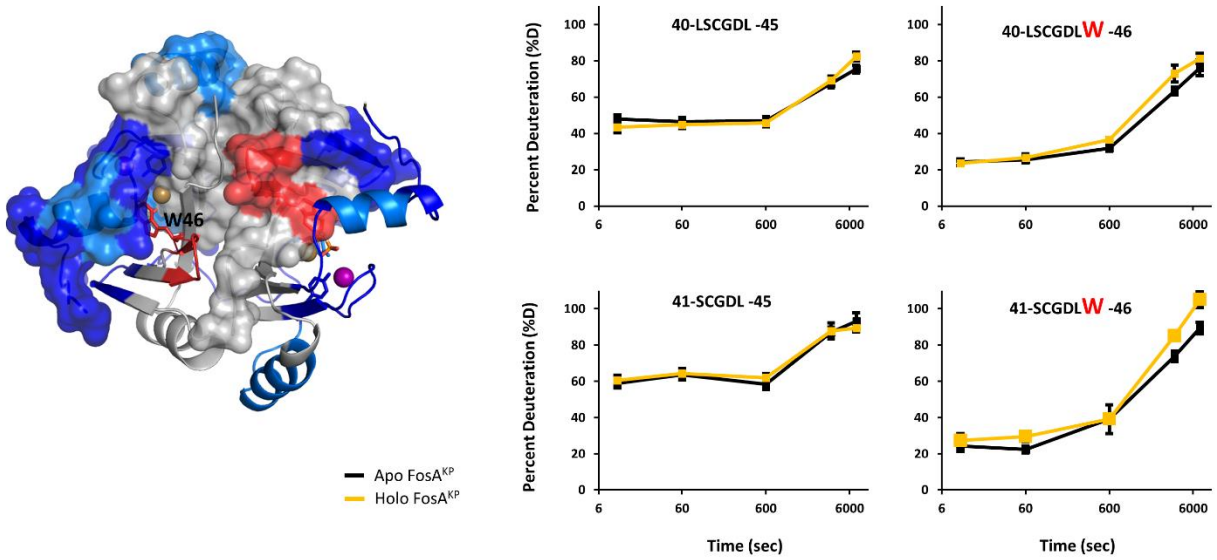

Figure S5. Fosfomycin-induced increase of hydrogen-deuterium exchange at W46. Peptides displaying significant differences in deuterium uptake are mapped on the crystal structure of $FosA^{KP}$. Regions with statistically significant decreases in deuterium at the earliest deuterium incubation time point (10 sec) are colored dark blue. Regions with decreases in deuterium uptake only observed at later time points are colored light blue. Regions with fosfomycin-induced increase in deuterium uptake are colored red. Important residues are shown as sticks, along with Mn^{2+} (gold), K^{+} (purple), and fosfomycin (sticks). Panel A through D show deuterium uptake traces for representative peptidic fragments in apo and holo $FosA^{KP}$. The sequences of the peptides are shown above each panels.