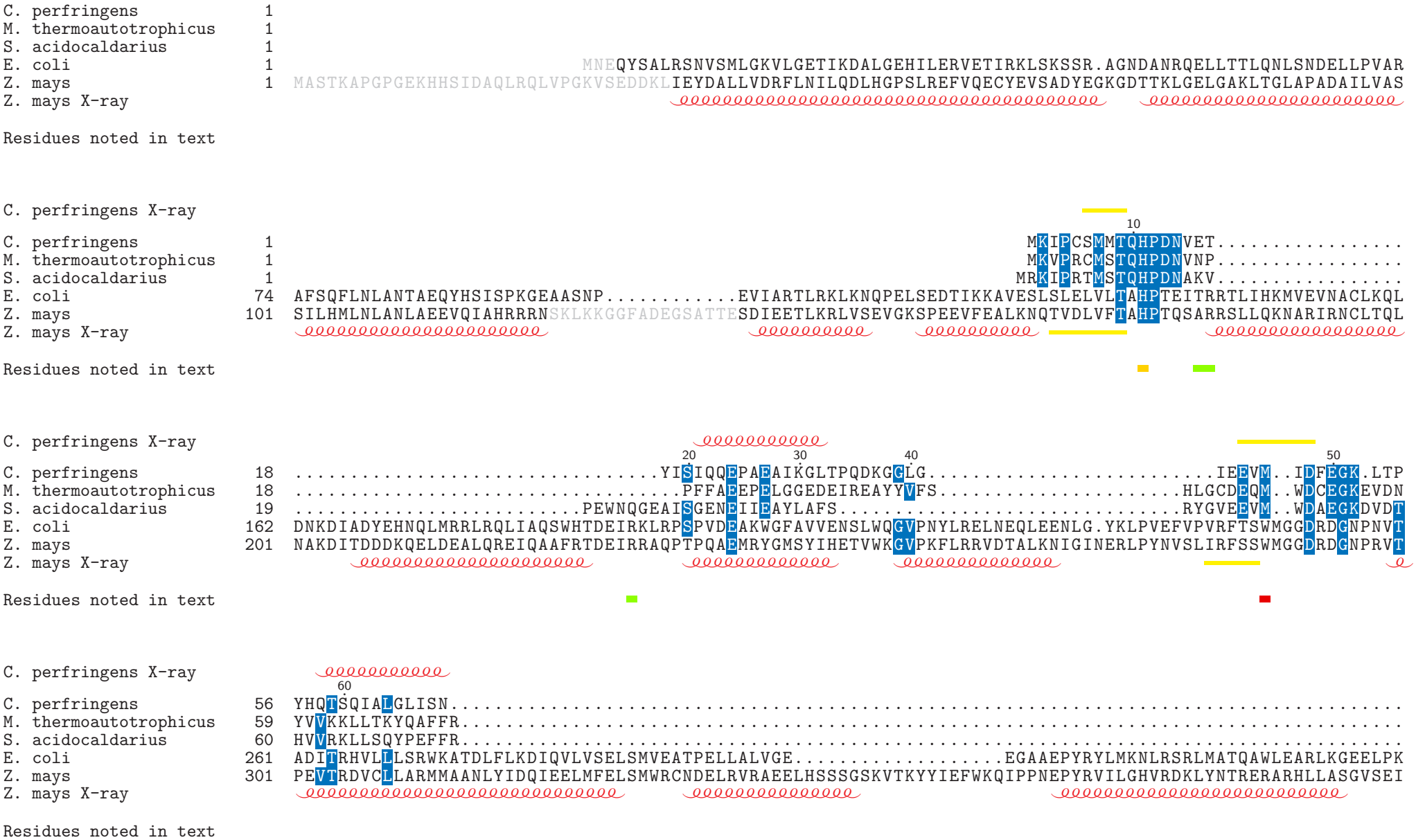
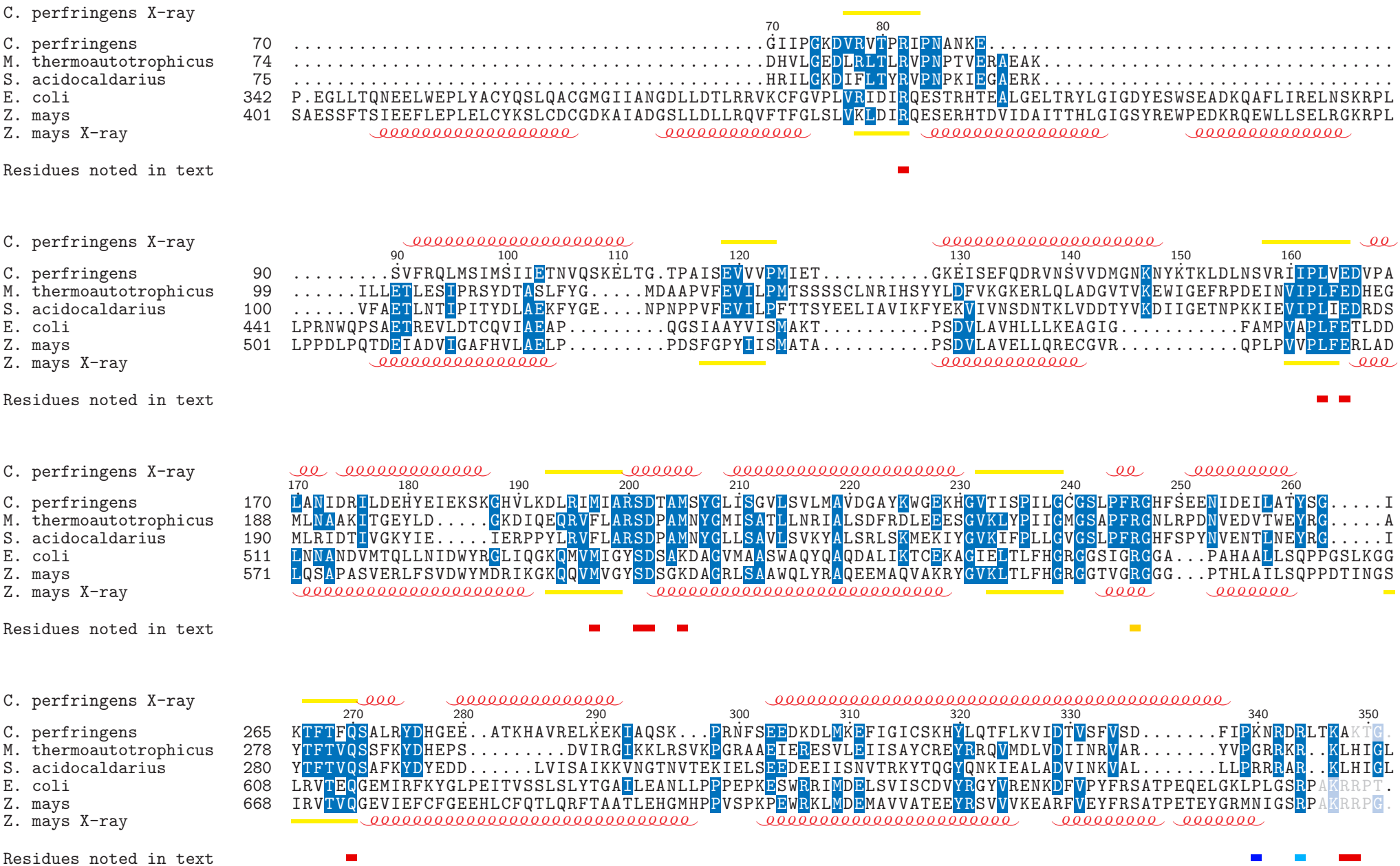
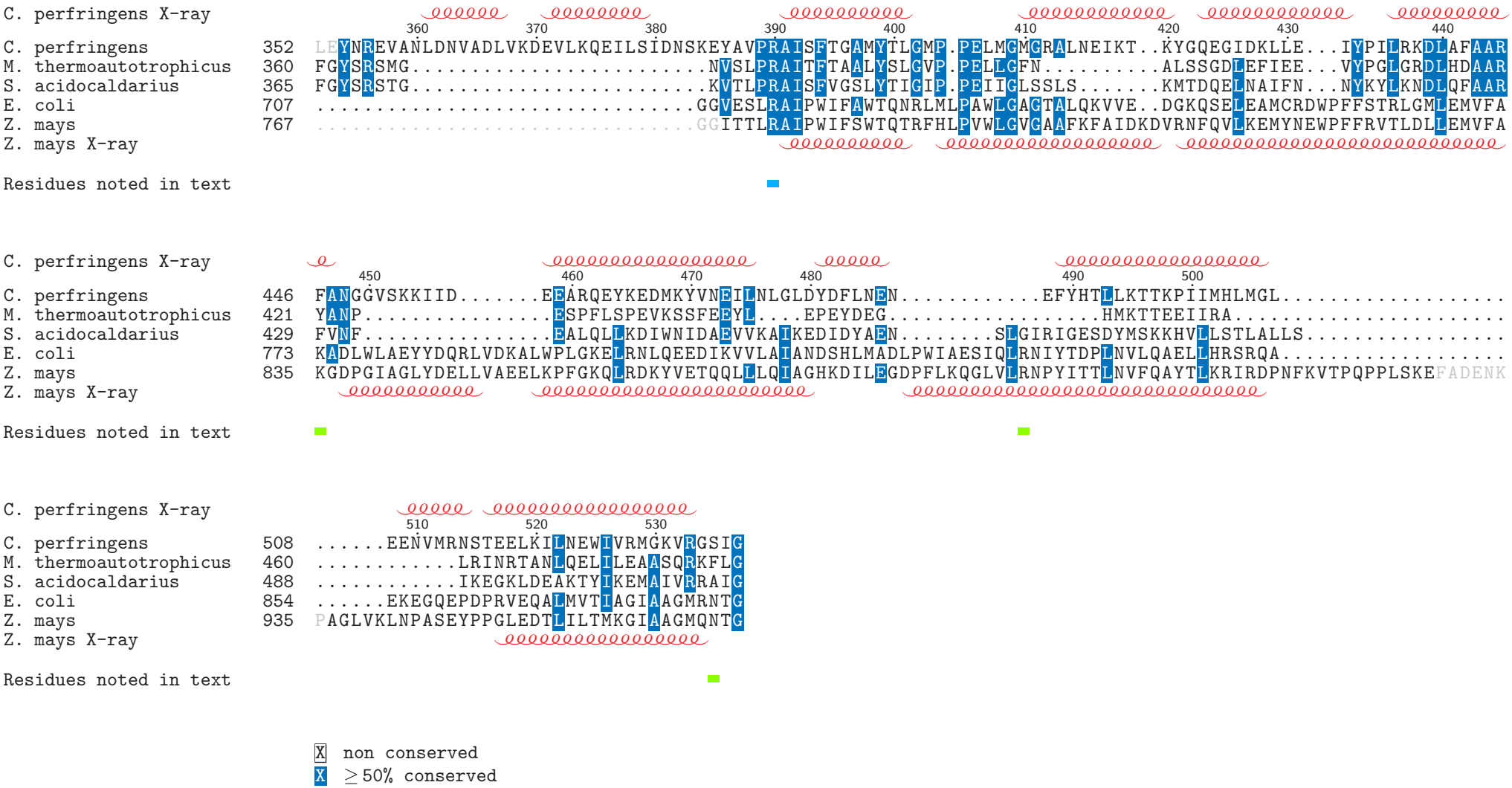
SUPPLEMENTARY FIGURE 1 A structure-based sequence alignment of PepcA with Pepc from *E. coli* (Protein Data Bank entry 1jqn) and *Z. mays* (1jqo) was generated with Strap.[1] Representative archaeal PepcA sequences were subsequently added to the alignment. The *Sulfolobus acidocaldarius* PepcA is sensitive to inhibition by aspartate; the *Methanothermobacter thermautotrophicus* PepcA is not. The secondary structure derived from the X-ray coordinates is shown for *C. perfringens* PepcA above the alignment, and for *Z. mays* Pepc below the alignment. Residues missing from the electron density are shown in lighter colors (for example the residues involved in HCO_3^- binding are missing from all three X-ray structures, at positions corresponding to 349-353 in the *C. perfringens* sequence). The insertion beginning at *C. perfringens* residue 360 is present only in PepcA sequences from *C. perfringens*, *Methanopyrus kandleri*, *Methanosarcina mazei*, *Methanosarcina barkeri* and *Methanosarcina acetivorans*. In the line highlighting residues discussed in the text, active-site residues are red; allosteric-site residues are green. His11 and Arg246 (His138 and Arg587 in *E. coli* numbering) in orange are in the active-site of the R-state; in the *E. coli* T-state His138 and Arg587 are not positioned in the active-site (Arg587 forms part of the allosteric inhibitor-binding site). Lys340 in dark blue is the residue in the proposed aspartate-binding site of PepcA responsible for determining the sensitivity of members of the PepcA family to inhibition by aspartate. Arg344 and Arg390 in light blue are in the active-site and are also part of the proposed aspartate-binding site in PepcA.

REFERENCES

1 Gille C, Frömmel C. STRAP: editor for STRuctural Alignments of Proteins. *Bioinformatics* 2001; **17**:377-8.


C. perfringens 1
M. thermoautotrophicus 1
S. acidocaldarius 1
E. coli 1 MNEQYSALRSNVSMLGKVLGETIKDALGEHILERVETIRKLSKSSR.AGNDANRQELLTTLQNLSNDELLPVAR
Z. mays 1 MASTKAPGPGEKHHSIDAQLRQLVPGKVSEDDKLIEYDALLVDRFLNILQDLHGPSLREFVQECYEVSADYEGKGDTTKLGELGAKLTGLAPADAILVAS
Z. mays X-ray
Residues noted in text
C. perfringens X-ray
10
C. perfringens 1 MKIPCSMMTQHPDNVET.....
M. thermoautotrophicus 1 MKVPRCMSTQHPDNVNP.....
S. acidocaldarius 1 MRKIPRTMSTQHPDNAKV.....
E. coli 74 AFSQFLNLANTAEQYHSISPKGEAASNP.....EVIARTLRKLKNQPELSEDTIKKAVESLSLELVLTAHPTEITRRTLIHKMVEVNACLKQL
Z. mays 101 SILHMLNLANLAEEVQIAHRRRNSKLKKGGFADEGSATTESDIEETLKRLVSEVGKSPEEVFEALKNQTVDLVFTAHPTQSARRSLLQKNARIRNCLTQL
Z. mays X-ray
Residues noted in text
C. perfringens X-ray
20 30 40 50
C. perfringens 18 .....YISIQQEPAEAIKGLTPQDKGGLG.....IEEVM..IDFEGK.LTP
M. thermoautotrophicus 18 .....PFFAEEPELGGEDEIREAYYVFS.....HLGCDEQM..WDCEGKEVDN
S. acidocaldarius 19 .....PEWNQGEAISGENEIIEAYLAFS.....RYGVEEVM..WDAEGKDVDT
E. coli 162 DNKDIADYEHNQLMRRLRQLIAQSWHTDEIRKLRPSPVDEAKWGFAVVENSLWQGVPNYLRELNEQLEENLG.YKLPVEFVPVRFTSWMGGDRDGNPNVT
Z. mays 201 NAKDITDDDKQELDEALQREIQAAFRTDEIRRAQPTPQAEMRYGMSYIHETVWKGVPKFLRRVDTALKNIGINERLPYNVSLIRFSSWMGGDRDGNPRVT
Z. mays X-ray
Residues noted in text
C. perfringens X-ray
60
C. perfringens 56 YHQTSQIALGLISN.....
M. thermoautotrophicus 59 YVVKKLLTKYQAFFR.....
S. acidocaldarius 60 HVVRKLLSQYPEFFR.....
E. coli 261 ADITRHVLLLSRWKATDLFLKDIQVLVSELSMVEATPELLALVGE.....EGAAEPYRYLMKNLRSRLMATQAWLEARLKGEELPK
Z. mays 301 PEVTRDVCLLARMMAANLYIDQIEELMFELSMWRCNDELRVRAEELHSSSGSKVTKYYIEFWKQIPPNEPYRVILGHVRDKLYNTRERARHLLASGVSEI
Z. mays X-ray
Residues noted in text


C. perfringens X-ray
C. perfringens 70
M. thermoautotrophicus 74
S. acidocaldarius 75
E. coli 342
Z. mays 401
Z. mays X-ray
Residues noted in text
C. perfringens X-ray
C. perfringens 90
M. thermoautotrophicus 99
S. acidocaldarius 100
E. coli 441
Z. mays 501
Z. mays X-ray
Residues noted in text
C. perfringens X-ray
C. perfringens 170
M. thermoautotrophicus 188
S. acidocaldarius 190
E. coli 511
Z. mays 571
Z. mays X-ray
Residues noted in text
C. perfringens X-ray
C. perfringens 265
M. thermoautotrophicus 278
S. acidocaldarius 280
E. coli 608
Z. mays 668
Z. mays X-ray
Residues noted in text


C. perfringens X-ray
C. perfringens 352 LEYNREVANLDNVADLVKDEVLKQEILSIDNSKEYAVPRAISFTGAMYTLGMP.PELMGMGRALNEIKT..KYGQEGIDKLLE...IYPILRKDLAFAAR
M. thermoautotrophicus 360 FGYSRSMG.....NVSLPRAITFTAALYSLGVP.PELLGFN.....ALSSGDLEFIEE...VYPGLGRDLHDAAR
S. acidocaldarius 365 FGYSRSTG.....KVTLPRAISFVGSLYTIGIP.PEIIGLSSLS.....KMTDQELNAIFN...NYKYLKNDLQFAAR
E. coli 707 .....GGVESLRAIPWIFAWTQNRLMLPAWLGAGTALQKVVE..DGKQSELEAMCRDWPFFSTRLGMLEMVFA
Z. mays 767 .....GGITTLRAIPWIFSWTQTRFHLPVWLGVGAAFKFAIDKDVRNFQVLKEMYNEWPFFRVTLDLLEMVFA
Z. mays X-ray
Residues noted in text
C. perfringens X-ray
C. perfringens 446 FANGGVSKKIID.....EEARQEYKEDMKYVNEILNLGLDYDFLNEN.....EFYHTLLKTTKPIIMHLMGL.....
M. thermoautotrophicus 421 YANP.....ESPFLSPEVKSSFEEYL....EPEYDEG.....HMKTTEEIIRA.....
S. acidocaldarius 429 FVNF.....EALQLLKDIWNIDAEVVKAIKEDIDYAEN.....SLGIRIGESDYMSKKHVLLSTLALLS.....
E. coli 773 KADLWLAEYYDQRLVDKALWPLGKELRNLQEEDIKVVLAIANDSHLMADLPWIAESIQLRNIYTDPLNVLQAELLHRSRQA.....
Z. mays 835 KGDPGIAGLYDELLVAEELKPFGKQLRDKYVETQQLLLQIAGHKDILEGDPFLKQGLVLRNPYITTLNVFQAYTLKRIRDPNFKVTPQPPLSKEFADENK
Z. mays X-ray
Residues noted in text
C. perfringens X-ray
C. perfringens 508 ......EENVMRNSTEELKILNEWIVRMGKVRGSIG
M. thermoautotrophicus 460 .....LRINRTANLQELILEAASQRKFLG
S. acidocaldarius 488 .....IKEGKLDEAKTYIKEMAIVRRAIG
E. coli 854 ......EKEGQEPDPRVEQALMVTIAGIAAGMRNTG
Z. mays 935 PAGLVKLNPASEYPPGLEDTLILTMKGIAAGMQNTG
Z. mays X-ray
Residues noted in text
X non conserved
X ≥ 50% conserved