|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Supplementary Table S2: Zinc-binding metalloenzymes** | | | | | | | | | | | | |
|  |  |  |  | **Gene** | | | | **Protein** | | | |  |
|  |  |  |  | **ZnCl2-MICs (µg/ml)** | | | |  | | | |  |
|  |  |  |  | **128** | **256** | | **512** |  |  |  |  |  |
| **Function** |  | **Symbol** | **Gene** | **n=7** | **n=136** | | **n=36** | **L** | **AV** | **C** | **I** | **REF** |
|  |  |  |  | **%** | **%** | | **%** | **AA** | **n** | **%** | **%** |  |
| Acetyl-CoA biosynthetic process | Acetate kinase | AckA | *ack*A | 100 | 100 | 100 | | 400 | 4 | 100 | 99.7 | [1] |
| Fatty acid biosynthesis process | Acyl carrier protein phosphodiesterase | AcpH | *acp*H | 100 | 100 | 100 | | 193 | 7 | 100 | 98.8 | [2] |
| DNA repair | Bifunctional transcriptional activator | Ada | *ada* | 100 | 100 | 100 | | 354 | 15 | 100 | 98.7 | [3] |
| Ethanol oxidation, formaldehyde catabolic process | S-(hydroxymethyl)glutathione dehydrogenase | FrmA | *adh*C (*frm*A) | 100 | 100 | 100 | | 369 | 5 | 100 | 98.8 | [4] |
| Alanyl-tRNA aminoacyla-tion (interpreta-tion of genetic code) | Alanyl-tRNA synthetase | AlaS | *ala*S | 100 | 100 | 100 | | 876 | 10 | 100 | 99.2 | [5] |
| Nitrogen metabolism | Allantoate amidohydrolase | AllC | *all*C | 57.1 | 96.3 | 100 | | 411 | 15 | 100 | 98.5 | [6] |
| Cell wall organisation | anhydro-N-acetylmuramoyl-l-alanine amidase | AmiD | *ami*D | 100 | 100 | 100 | | 276 | 8 | 100 | 99.2 | [7] |
| Cell wall organisation | N-acetyl-anhydromuramyl-L-alanine amidase | AmpD | *amp*D | 100 | 100 | 100 | | 183 | 8 | 100 | 99.7 | [8] |
| L-arginine biosynthesis | acetylornithine deacetylase | ArgE | *arg*E | 100 | 100 | 100 | | 383 | 7 | 100 | 99.1 | [9] |
| Carbon utilization | carbonic anhydrase | Can | *can* | 100 | 100 | 100 | | 220 | 1 | 100 | 100 | [10] |
| Cytidine deamination | Cytidine deaminase | Cdd | *cdd* | 100 | 100 | 100 | | 294 | 5 | 100 | 99.8 | [11] |
| Carbon utilization | Carbonic anhydrase | CynT | *cyn*T | 100 | 96.3 | 97.2 | | 219 | 6 | 100 | 99.7 | [12] |
| Cysteinyl-tRNA aminoacyla-tion (interpretion of genetic code) | Cysteinyl-tRNA synthetase | CysS | *cys*S | 100 | 100 | 100 | | 461 | 6 | 100 | 99.8 | [13] |
| L-lysine biosynthesis via DAP pathway (bacterial cell wall) | N-Succinyl-L-diaminopimelate desuccinylase | DapE | *dap*E | 100 | 100 | 100 | | 375 | 10 | 100 | 99.9 | [14] |
| DNA replication (synthesis of RNA primer) | ssDNA-dependent RNA-polymerase | DnaG | *dna*G | 100 | 100 | 100 | | 581 | 6 | 100 | 99.9 | [15] |
| Repair, DNA replication | Chaperone Hsp70 (heat shock 70 kDa) | Dank | *dna*K | 100 | 100 | 100 | | 638 | 4 | 100 | 99.5 | [1] |
| dUMP biosynthesis | Deoxyuridine triphosphatase (dUTPase) | Dut | *dut* | 100 | 100 | 100 | | 151 | 2 | 100 | 99.8 | [16] |
| rRNA processing | Zinc phosphodiesterase | Rbn | *ela*C (*rbn*) | 100 | 100 | 100 | | 305 | 8 | 100 | 99.2 | [17] |
| Gluconeogenesis and glycolytic process | Fructose-bisposphate aldolase, class II | FbaA | *fba*A | 100 | 100 | 100 | | 359 | 2 | 100 | 99.98 | [18] |
| One-carbon metabolic process | GTP cyclohydrolase | FolE | *fol*E | 100 | 100 | 100 | | 222 | 2 | 100 | 99.98 | [19] |
| D-arabinose and L-fucose catabolic process | L-fuculose-phosphate aldolase & D-ribulose-phosphate aldolase | FucA | *fuc*A | 100 | 100 | 100 | | 215 | 4 | 100 | 99.4 | [20] |
| Galactose metabolism | Galactose-1-phosphate uridylyltransferase | GalT | *gal*T | 100 | 100 | 100 | | 348 | 13 | 100 | 98.8 | [21] |
| Methylgly-oxal catabolic process to D-lactate via S-lactoyl-glutathione | Hydroxyacylglutathione hydrolase | GloB | *glo*B | 100 | 100 | 100 | | 251 | 11 | 100 | 99.1 | [22] |
| Glutamyl-tRNA aminoacyla-tion | Glutamyl-tRNA-synthetase | GltX | *glt*X | 100 | 100 | 100 | | 471 | 4 | 100 | 99.4 | [23] |
| Glycine biosynthetic process from serine | Serine hydroxmethyltransferase (SHMT) | GlyA | *gly*A | 100 | 100 | 100 | | 417 | 1 | 100 | 100 | [1] |
| Guanine catabolic process | Guanine deaminase | GuaD | *gua*D | 100 | 99.3 | 100 | | 439 | 8 | 100 | 99.4 | [24] |
| Heme biosynthetic process | Porphobilinogen synthase | HemB | *hem*B | 100 | 100 | 100 | | 324 | 2 | 100 | 99.96 | [25] |
| Proteolysis | Integral membrane ATP-dependent zinc metallopeptidase | FtsH | *hfl*B (*fts*H) | 100 | 100 | 100 | | 644 | 1 | 100 | 100 | [26] |
| Cellular protein modification process | Hydrogenase maturation regulator | HypA | *hyp*A | 100 | 100 | 100 | | 116 | 3 | 100 | 99.9 | [27] |
| Cellular protein modification process | **I**so**a**spartyl **d**ipeptidase | IadA | *iad*A | 100 | 100 | 100 | | 390 | 18 | 100 | 99.4 | [28] |
| Cellular response to DNA damage stimulus | Isopentenyl diphosphate isomerase | Idi | *idi* | 100 | 100 | 100 | | 182 | 10 | 100 | 97.9 | [29] |
| Isoleucyl-tRNA aminoacyla-tion | Isoleucyl-tRNA synthetase | IleS | *ile*S | 100 | 100 | 100 | | 938 | 14 | 100 | 99.0 | [30] |
| Lipid A biosynthesis | UDP-3-O-acyl-N-acetylglucosamine deacetylase | LpxC | *lpx*C | 100 | 100 | 100 | | 305 | 2 | 100 | 99.99 | [31] |
| Lysyl-tRNA aminoacyla-tion | Lysine-tRNA ligase | LysU | *lys*U | 100 | 100 | 100 | | 505 | 7 | 100 | 99.2 | [32] |
| cell wall manno-protein biosynthetic process | mannose-6-phophate isomerase | ManA | *man*A | 100 | 100 | 100 | | 391 | 10 | 100 | 99.0 | [33] |
| Methionine biosynthetic process | homocysteine transmethylase | MetE | *met*E | 100 | 100 | 100 | | 753 | 11 | 100 | 99.4 | [34] |
| Methionyl-tRNA aminoacyla-tion | Methionyl-tRNA synthetase | MetG | *met*G | 100 | 100 | 100 | | 677 | 11 | 100 | 99.7 | [35] |
| DNA repair (base-excision repair) | Formamidopyrimidine DNA glycosylase | MutM | *mut*M | 100 | 100 | 100 | | 269 | 5 | 100 | 99.6 | [36] |
| Carbohydrate metabolic process | **N**-**A**cetyl**g**lucosamine-6-phosphate deacetylase | NagA | *nag*A | 100 | 100 | 100 | | 382 | 10 | 100 | 99.2 | [37] |
| DNA repair (base-excision repair) | Endonuclease VIII | Nei | *nei* | 100 | 100 | 100 | | 263 | 12 | 100 | 98.3 | [38] |
| Pyridoxine biosynthetic process | 4-Hydroxy-L-threonine phosphate dehydrogenase | PdxA | *pdx*A | 100 | 100 | 100 | | 329 | 12 | 100 | 98.3 | [39] |
| Pyridoxine biosynthetic process | Pyridoxal kinase | PdxK | *pdx*K | 100 | 100 | 100 | | 283 | 5 | 100 | 99.4 | [40] |
| Peptide catabolic process | Cytosol aminopeptidase | PepA | *pep*A | 100 | 100 | 100 | | 503 | 3 | 100 | 99.99 | [41] |
| Dephospho-rylation | Alkaline phosphatase | PhoA | *pho*A | 100 | 100 | 100 | | 471 | 16 | 100 | 98.5 | [42] |
| DNA repair (base-excision repair) | DNA polymerase I | PolA | *pol*A | 100 | 100 | 100 | | 928 | 13 | 100 | 99.4 | [43] |
| UMP biosynthetic process | Aspartate carbomoyltransferase | PyrB | *pyr*B | 100 | 100 | 100 | | 311 | 6 | 100 | 99.1 | [44] |
| UMP biosynthetic process | Dihydroorotase | PyrC | *pyr*C | 100 | 100 | 100 | | 348 | 8 | 100 | 99.0 | [45] |
| Pyrimidine nucleotide biosynthetic process | Aspartat carbamoyltransferase regulatory chain | PyrI | *pyr*I | 100 | 100 | 100 | | 153 | 2 | 100 | 99.96 | [46] |
| DNA repair | DNA helicase | RecQ | *rec*Q | 100 | 100 | 100 | | 609 | 9 | 100 | 98.6 | [47] |
| Rhamnose catabolic process | Rhamnulose-1-phosphate aldolase | RhaD | *rha*D | 100 | 100 | 100 | | 274 | 13 | 100 | 96.9 | [48] |
| Translation | Ribosomal protein L13 | RplM | *rpl*M | 100 | 100 | 100 | | 142 | 1 | 100 | 100 | [1] |
| Transcription | RNA polymerase β-subunit | RpoB | *rpo*B | 100 | 100 | 100 | | 1342 | 2 | 100 | 99.99 | [49] |
| Transcription | RNA polymerase subunit β’ | RpoC | *rpo*C | 100 | 100 | 100 | | 1407 | 3 | 100 | 99.98 | [50] |
| Regulation of translation (mRNA polyadenylation) | Ribosomal protein S15 | RpsO | *rps*O | 100 | 100 | 100 | | 89 | 2 | 100 | 99.9 | [1] |
| Translation (ribosomal small subunit assembly) | Ribosomal protein S16 | RpsP | *rps*P | 100 | 100 | 100 | | 82 | 1 | 100 | 100 | [1] |
| Translation (ribosomal small subunit assembly) | Ribosomal protein S17 | RpsQ | *rps*Q | 100 | 100 | 100 | | 84 | 1 | 100 | 100 | [1] |
| rRNA base methylation | Methyltransferase | RlmA | *rrm*A (*rlm*A) | 100 | 100 | 100 | | 269 | 13 | 100 | 99.1 | [51] |
| Superoxide metabolic process | Superoxide dismutase | SodC | *sod*C | 100 | 100 | 100 | | 173 | 5 | 100 | 99.5 | [52] |
| tRNA wobble adenosine to inosine editing | **t**RNA **a**denosine **d**eaminase | TadA | *tad*A | 100 | 100 | 100 | | 167 | 4 | 100 | 99.4 | [53] |
| L-serine/L-threonine catabolic process | Threonine dehydrogenase | Tdh | *tdh* | 100 | 100 | 100 | | 341 | 6 | 100 | 96.6 | [54] |
| Queuosine biosynthetic process | Queuine tRNA-ribosyltransferase | Tgt | *tgt* | 100 | 100 | 100 | | 375 | 4 | 100 | 99.6 | [55] |
| Regulation of translation (tRNA aminoacylation) | Threonyl-tRNA synthetase | ThrS | *thr*S | 100 | 100 | 100 | | 642 | 6 | 100 | 99.5 | [56] |
| Cellular response to antibiotic | Transketolase 1 | TktA | *tkt*A | 100 | 100 | 100 | | 663 | 7 | 100 | 98.8 | [57] |
| Pentose-phosphate shunt | Transketolase 2 | TktB | *tkt*B | 100 | 100 | 100 | | 667 | 14 | 100 | 99.0 | [1] |
| DNA topological change | DNA topoisomerase | TopA | *top*A | 100 | 100 | 100 | | 865 | 8 | 100 | 99.5 | [58] |
| Elongation | Translation elongation factor | Tsf | *tsf* | 100 | 100 | 100 | | 283 | 3 | 100 | 99.8 | [1] |
| DNA repair | Excision nuclease subunit A | UvrA | *uvr*A | 100 | 100 | 100 | | 940 | 5 | 100 | 99.98 | [59] |
| Mannitol metabolic process | D-mannonate oxidoreductase | UxuB | *uxu*B | 100 | 100 | 100 | | 486 | 15 | 100 | 98.1 | [60] |
| Bacterial-type flagellum-dependent swarming motility | Probable phosphatase | YcdX | *ycd*X | 100 | 100 | 100 | | 245 | 6 | 100 | 99.1 | [61] |

All 179 porcine commensal *E. coli* were screened with respect to the presence or absence of 69 factors involved in zinc homeostasis as zinc-binding metalloenzymes. Predicted amino acid sequence lengths were compared to those of *E. coli* K-12 MG1655 in order to check for putative premature stop codons or deletions affecting the putative function of the protein. None of the amino acid sequence variations (AV) was solely associated with a particular ZnCl2 MIC. Amino acid sequence variation among the isolate collection primarily reflects the isolate’s genetic backgrounds.

**Abbreviations:**

n, number of isolates; a, number of isolates with particular ZnCl2 MIC; L, length; AA, amino acid sequence; AV, number of amino acid sequence variants; C, amino acid sequence coverage with respect to reference protein in *E. coli* K-12 MG1655; I, maximum amino acid sequence identity among the 179 isolates;

**References**

[1] (Katayama et al., 2002); [2] (Vagelos and Larrabee, 1967); [3] (Mielecki and Grzesiuk, 2014); [4] (Gutheil et al., 1997); [5] (Sood et al., 1999); [6] (Agarwal et al., 2007); [7] (Kerff et al., 2010); [8] (Jacobs et al., 1994); [9] (Javid-Majd and Blanchard, 2000); [10] (Merlin et al., 2003); [11] (Xiang et al., 1997); [12] (Guilloton et al., 1992); [13] (Newberry et al., 2002); [14] (Lin et al., 1988); [15] (Loscha et al., 2004); [16] (Larsson et al., 1996); [17] (Kostelecky et al., 2006); [18] (Labbe et al., 2011); [19] (Auerbach et al., 2000); [20] (Dreyer and Schulz, 1996); [21] (Geeganage and Frey, 1999); [22] (O'Young et al., 2007); [23] (Banerjee et al., 2004)[24] (Maynes et al., 2000); [25] (Jaffe et al., 2002); [26] (Tomoyasu et al., 1995); [27] (Douglas et al., 2013) [28](Martí-Arbona et al., 2005); [29] (Lee and Poulter, 2006); [30] (Xu et al., 1994); [31] (Gattis et al., 2010); [32] (Chen et al., 2013); [33] (Gao et al., 2005); [34] (Hondorp and Matthews, 2004); [35] (Mayaux and Blanquet, 1981); [36] (Boiteux et al., 1990); [37] (Hall et al., 2007); [38] (Kropachev et al., 2006); [39] (Sivaraman et al., 2003); [40] (di Salvo et al., 2004); [41] (Straeter and Lipscomb, 1995); [42] (Sowadski et al., 1985); [43] (Springgate et al., 1973); [44] (Lipscomb and Kantrowitz, 2012); [45] (Lee et al., 2005); [46] (Rastogi et al., 1998); [47] (Liu et al., 2004); [48] (Kroemer et al., 2003); [49] (Alifano et al., 2015); [50] (Morse et al., 1996); [51] (Das et al., 2004); [52] (Pesce et al., 1997); [53] (Kim et al., 2006); [54] (Johnson et al., 1998); [55] (Chong et al., 1995); [56] (Caillet et al., 2007); [57] (Domain et al., 2007); [58] (Lu et al., 2011); [59] (Wang et al., 1994); [60] (Nguyen et al., 2008); [61](Teplyakov et al., 2003)

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