SUPPLEMENT

**Table S1.** Primers used for the amplification of gene fragments from the corresponding genomic DNAs

| Uniprot ID | Locus tag | Organism | Primer sequence, 5′ to 3′ |
|---|---|---|---|
| B9JKN9 | *arad_9439* | *Agrobacterium radiobacter* K84 | gtcctccggcgc**catatg**ccgggggccg<br>ctccgtttatcg**ctcgag**cgtttcgtcgc |
| Q92TT0 | *sm_b20710* | *Rhizobium meliloti* 1021 | cagaggaggcaat**catatg**acgaaatcgaac<br>caaaaggcgctcgc**ggatcc**gttcactcgag |
| Q120Q9 | *bpro_5116* | *Polaromonas* sp. JS666 | gaaggaaatc**catatg**aagaaaatcggaatg<br>cgattcatcatg**ctcgag**ctaggcttctg |
| Q12H32 | *bpro_0195* | *Polaromonas* sp. JS666 | caggagaca**catatg**aaacacattggaatg<br>ctgtagcgcgccaagg**ctcgag**ttcactgc |
| D7A6R3 | *snov_2985* | *Starkeya novella* ATCC 8093 | gaaaacgaggacaag**catatg**gcgaaggttg<br>gaatgcaatatcgg**ctcgag**atcacgcatc |
| Q1LMW1 | *rmet_1632* | *Ralstonia metallidurans* CH34 | cttcaaccggatatc**catatg**agcaagatc<br>gtcggagttaagt**ctcgag**atcagcctttg |
| B9J8U1 | *arad_3557* | *Agrobacterium radiobacter* K84 | gggagggcagct**catatg**accggtcggaag<br>cagacgacgttt**ggatcc**gctgttgtcgctc |
| A9CHF6 | *atu2737* | *Agrobacterium fabrum* C58 | ggaggttcact**catatg**agtgtcggcgggg<br>cttgtcgtcgct**ctcgag**atcgcctcagcc |
| Q1QSM4 | *csal_3190* | *Chromohalobacter salexigens* DSM 3043 | gaggatgataac**catatg**aatgacaatcgag<br>ccgttttggcg**ggatcc**gcttaaccgtc |
| Q1QSN6 | *csal_3178* | *Chromohalobacter salexigens* DSM 3043 | gacgaggaacgc**gagatg**acacaagcacg<br>ccgatgatgg**ggatcc**tcattcctcctc |
| B9J8U2 | *arad_3558* | *Agrobacterium radiobacter* K84 | gcgagggtgaa**catatg**catatcgccattatc<br>catcaatccggt**ctcgag**gaaagcgatcttc |
| A9CHF5 | *atu2738* | *Agrobacterium fabrum* C58 | gatagaggaggaa**catatg**catatcgcaatc<br>ccgcccatcagg**ctcgag**ccgataaaggcg |
| Q1QSM2 | *csal_3192* | *Chromohalobacter salexigens* DSM 3043 | gaaaggaatga**catatg**cacgtactgataac<br>gggccgatggcg**aagctt**cattgaggcggc |
| Q1QSN8 | *csal_3176* | *Chromohalobacter salexigens* DSM 3043 | ccggggagaca**catatg**caagtcatcgtg<br>cgccgccgggcc**aagctt**ggcgtcaggtgc |
| B9JKP0 | *arad_9440* | *Agrobacterium radiobacter* K84 | cggagacaatc**catatg**cccgtctttgcgg<br>gtcggcaatcgc**ctcgag**caagagtgtcatc |

| | | | |
|---|---|---|---|
| B9JKP1 | *arad_9441* | *Agrobacterium radiobacter* K84 | caaaaggataaa**catatg**acactcttgctc ggtgtccctac**ctcgag**tgcctaatgcatc |
| Q1LQ56 | *rmet_0834* | *Cupriavidus metallidurans* ATCC 43123 | ccggagacaacc**catatg**aatgccgcttcc gccccccgccacg**aagctt**ggcccgtcagcg |
| Q1LMW0 | *rmet_1633* | *Cupriavidus metallidurans* ATCC 43123 | ccaggagtaac**catatg**acccaacatctgc gatcttgctcat**ctcgag**atccggttgaag |
| Q1LMV9 | *rmet_1634* | *Cupriavidus metallidurans* ATCC 43123 | ctggagta**catatg**agtgaacaccgtattc gtcatgcggtta**ctcgag**gttcaatcggtc |

Note: Restriction endonuclease sites are highlighted in bold.

**Table S2.** Aldonic sugar acid library used for the screening the substrate specificity of ThrDH and EryDH homologs activity screening (34 monocarboxylic sugar acids, including three branched sugar acids, such as D-apionate, (*R*)-pantoate, and 2,3-dihydroxyisovalerate)

| **3C-Sugar acids** | **5C-Sugar acids** | **6C-Sugar acids** | **6C-Sugar acids** | **Others** |
|---|---|---|---|---|
| L-lactic acid | D-arabinonic acid | D-altronic acid | D-gulonic acid | D-fuconic acid |
| D-glyceric acid | L-arabinonic acid | L-altronic acid | L-gulonic acid | L-fuconic acid |
| L-glyceric acid | D-lyxonic acid | D-allonic acid | D-idonic acid | D-rhamnonic acid |
| **4C-Sugar acids** | L-lyxonic acid | L-allonic acid | L-idonic acid | 6-deoxy-L-talonic acid |
| D-erythronic acid | D-ribonic acid | D-galactonic acid | D-mannonic acid | D-apionate |
| L-erythronic acid | L-ribonic aid | L-galactonic acid | L-mannonic acid | (*R*)-pantoate |
| D-threonic acid | D-xylonic acid | D-gluconic acid | D-talonic acid | 2,3-dihydroxyisovalerate |

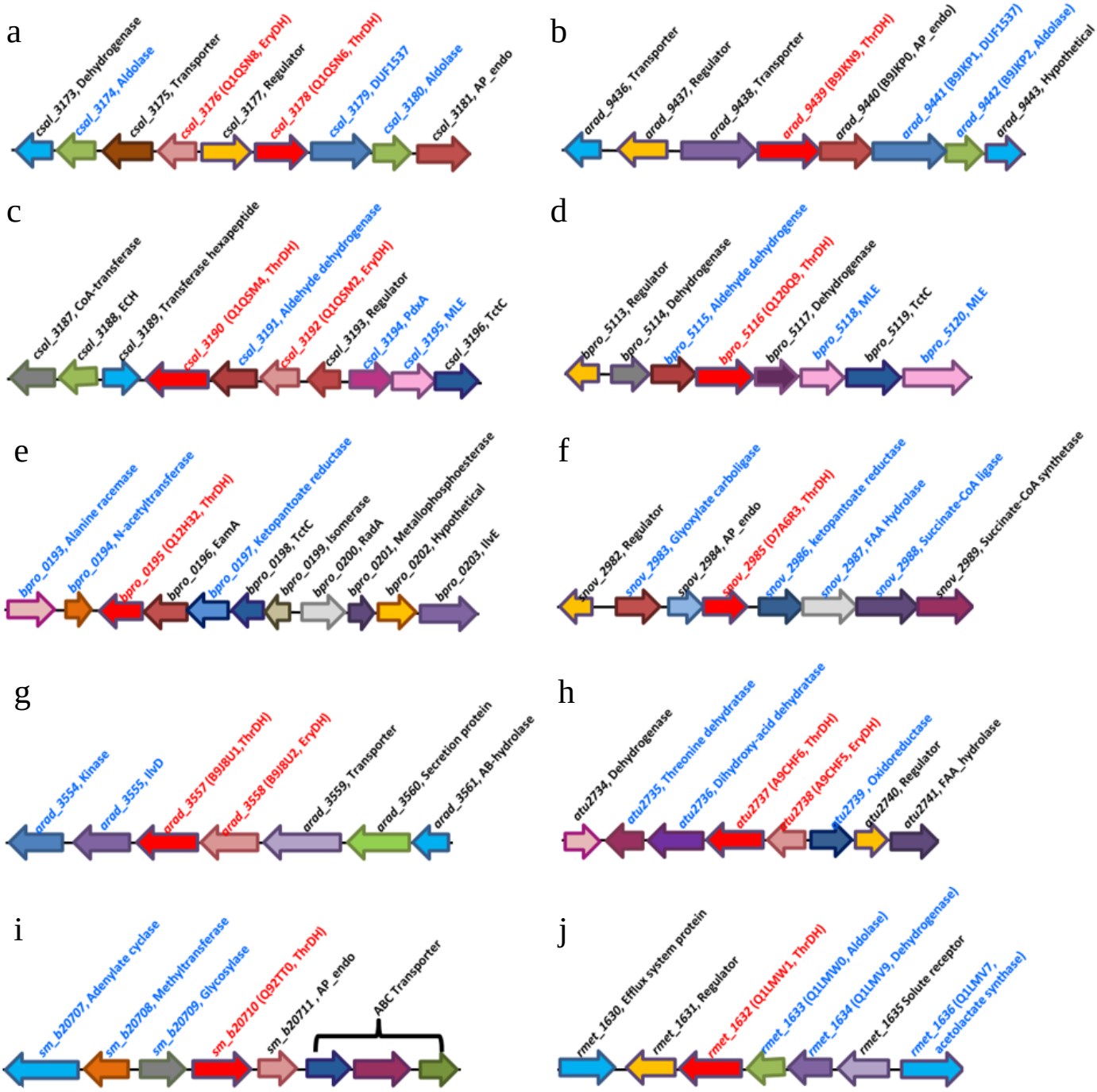


**Fig. S1.** Genome contexts for the ThrDH homologs studied in this work; the tested enzymes are highlighted in red; neighboring distinctive functional genes are highlighted in blue. Q1QSN6 and B9JKN9 have the DUF1537/aldolase gene context (a, b); Q1QSM4 and Q120Q9 have the PdxA/MLE gene context (c, d); and Q12H32 and D7A6R3 have uncharacterized neighboring keto pantoate reductase (e, f), while several unknown dehydratases reside near B9J8U1 and A9CHF6 (g, h).

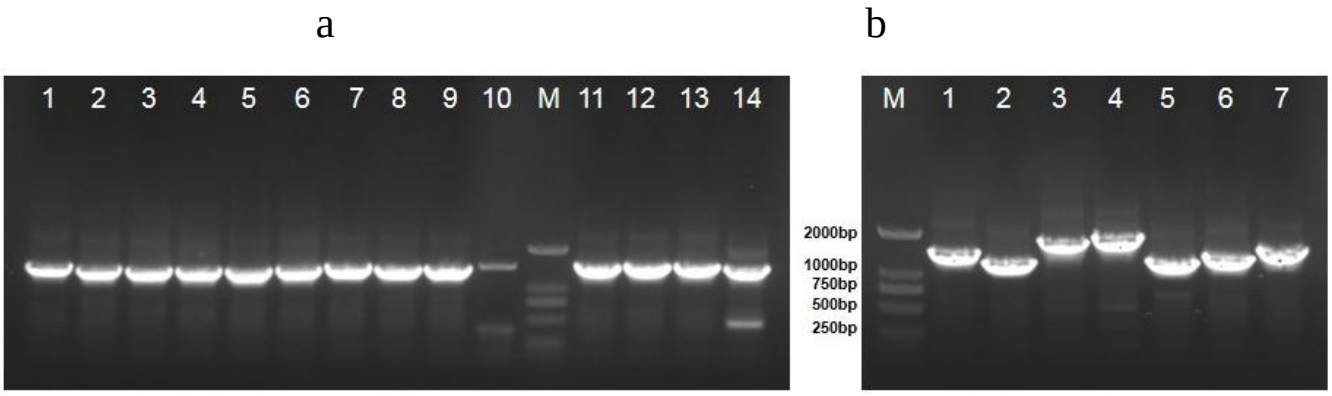


**Fig. S2.** a) Electrophoresis of gene fragments of 10 ThrDH homologs (lanes 1-10) and 4 EryDH homologs (lanes 11-14) used for cloning. Lanes: 1) B9JKN9; 2) Q92TT0; 3) Q120Q9; 4) Q12H32; 5) D7A6R3; 6) Q1LMW1; 7) B9J8U1; 8) A9CHF6; 9) Q1QSM4; 10) Q1QSN6; 11) B9J8U2; 12) A9CHF5; 13) Q1QSM2; 14) Q1QSN8; M) DNA markers. b) Fragments of genes involved in two catabolic pathways. Lanes: 1) B9JKN9; 2) B9JKP0; 3) B9JKP1; 4) Q1LQ56; 5) Q1LMW0; 6) Q1LMW1; 7) Q1LMV9. Unless specified, all genes were cloned into the pET-28a plasmids.

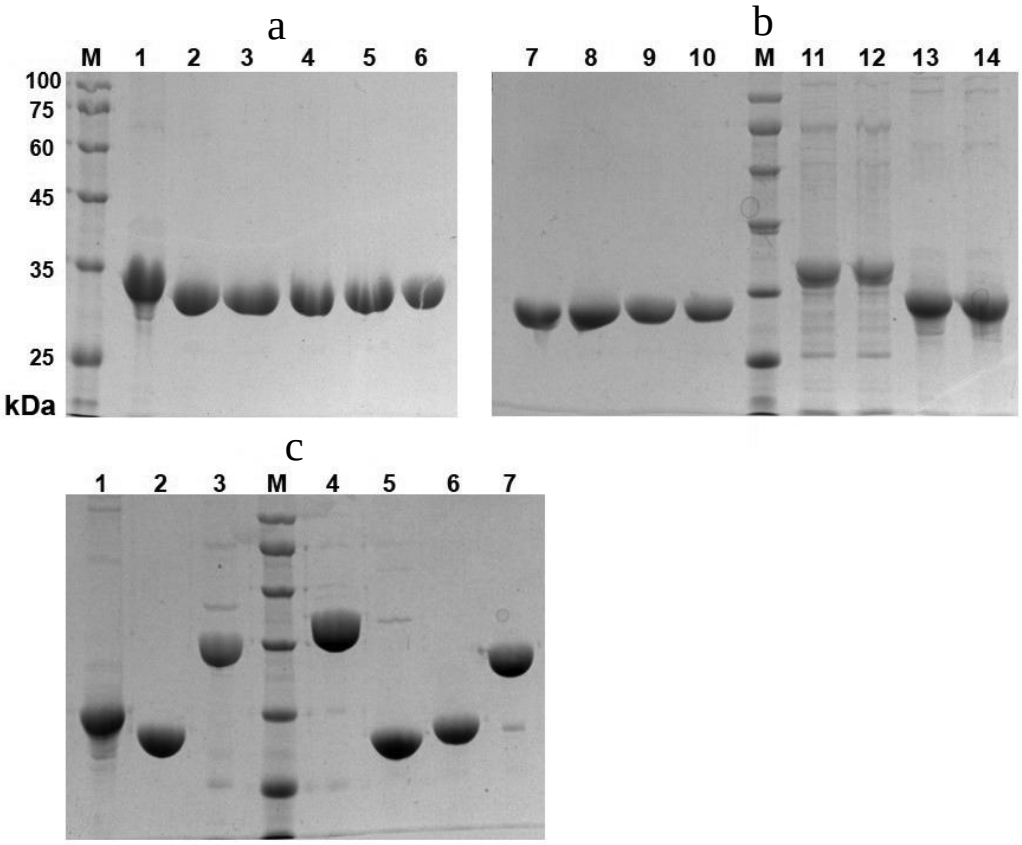


**Fig. S3.** SDS-PAGE (12%) of purified enzymes. a, b) Ten ThrDH homologs (lanes 1-10) and four EryDH homologs (lanes 11-14). Lanes: 1) B9JKN9; 2) Q92TT0; 3) Q120Q9; 4) Q12H32; 5) D7A6R3; 6) Q1LMW1; 7) B9J8U1; 8) A9CHF6; 9) Q1QSM4; 10) Q1QSN6; 11) B9J8U2; 12) A9CHF5; 13) Q1QSM2; 14) Q1QSN8; M) protein markers. c) Catabolic enzymes related to ThrDH. Lanes: 1) B9JKN9; 2) B9JKP0; 3) B9JKP1; 4) Q1LQ56; 5) Q1LMW0; 6) Q1LMW1; 7) Q1LMV9.