Conservation of a glutamate residue in ATP-citrate lyase and succinyl-CoA synthetase

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Succinyl-CoA synthetase (SCS), the enzyme that catalyzes the only substrate-level phosphorylation in the citrate cycle, is the prototype for a family of ADP- or GDP-forming acyl-CoA synthetases that includes ATP-citrate lyase (ACLY) [1]. These enzymes catalyze the formation of a thioester bond between an organic acid and CoA, using the energy of nucleotide triphosphate (NTP) and in the presence of magnesium ions. A histidine residue is transiently phosphorylated during catalysis [2], leading to the proposed catalytic mechanism:

$$E + NTP \rightleftharpoons E\text{–}PO_3 + NDP \quad (1)$$

$$E\text{–}PO_3 + carboxylate \rightleftharpoons E\cdot carboxyl\text{–}phosphate \quad (2)$$

$$E\cdot carboxyl\text{–}phosphate + CoA \rightleftharpoons E + carboxyl\text{–}CoA + P_i \quad (3)$$

where E represents the enzyme; –, a covalent bond; and · , noncovalent interactions. For SCS, the carboxylate is succinate; for ACLY, it is citrate and there is fourth step in which citryl-CoA is cleaved to form acetyl-CoA and oxaloacetate.

A glutamate residue of ACLY, E599, was proposed to play a role in the cleavage of citryl-CoA [3]. This glutamate residue is conserved not only in ACLYs but also in SCSs (Fig. 1). The structures of SCSs and ACLYs found in the Protein Data Bank [4] are used to investigate the role of this conserved glutamate residue.

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Human ACLY                      IRTIAIIAEGIPEALTRKLIKKA-DQKGVTIIGPATVGGIKPGCFKIGNTGGMLDNILASKLYR
Chlorobium limicola ACLY        IQLVSMITEGVPEKDAKRLKKLA-QKLGKMLNGPSSIGIMSAGECRLGVIGGEFKNLKLCNLYR
Human GTPSCS α-subunit          IPLVVCITEGIPQQDMVRVKHKLLRQEKTRLIGPNCPGVINPGECKIGIMPG--------HIHK
Escherichia coli α-subunit      IKLIITITEGIPTLDMLTVKVKL-DEAGVRMIGPNCPGVITPGECKIGIQPG--------HIHK
Thermus aquaticus α-subunit     IPLIVLITEGIPTLDMVRAVEEI-KALGSRLIGGNCPGIISAEETKIGIMPG--------HVFK
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Figure 1. Alignment of portions of the sequences of ACLYs and SCSs. The alignment shows conservation of a glutamate residue, E599 in human ACLY, E112 in the A-subunit of Chlorobium limicola ACLY, E105α of human GTPSCS, E98α of E. coli SCS, and E97α of Thermus aquaticus GTPSCS.

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