SUPPORTING INFORMATION

# Human 2-Oxoglutarate Dehydrogenase Complex E1 Component Forms a Thiamin-derived Radical by Aerobic Oxidation of the Enamine Intermediate.

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**SI Table S1**. Peptic peptides originated from E1o-h [a]

| Peptides | | | Monoisotopic Mass $[M+H]^+$ (Da) | | Error (ppm) |
|---|---|---|---|---|---|
| **No.** | **Position** | **Sequence** | **Experimental** | **Theoretical** | |
| 1 | 2-8 | SAPVAAE | 644.3248 | 644.3250 | -0.2 |
| 2 | 9-17 | PFLSGTSSN | 909.4309 | 909.4312 | -0.3 |
| 3 | 18-22 | YVEEM | 670.2756 | 670.2753 | 0.5 |
| 4 | 22-32 | MYCAWLENPKS | 1341.5956 | 1341.5966 | -0.8 |
| 5 | 34-47 | HKSWDIFFRNTNAG | 1692.8260 | 1692.8241 | 1.1 |
| 6 | 41-65 | FRNTNAGAPPGTAYQSPLPLSRGSL | 2572.3259 | 2572.3267 | -0.3 |
| 7 | 66-74 | AAVAHAQSL | 867.4678 | 867.4683 | -0.5 |
| 8 | 75-84 | VEAQPNVDKL | 1112.5938 | 1112.5946 | -0.7 |
| 9 | 85-89 | VEDHL | 612.2986 | 612.2988 | -0.2 |
| 10 | 90-94 | AVQSL | 517.2980 | 517.2980 | 0.0 |
| 11 | 95-114 | IRAYQIRGHHVAQLDPLGIL | 2270.2893 | 2270.2880 | 0.6 |
| 12 | 115-135 | DADLDSSVPADIISSTDKLGF | 2166.0460 | 2166.0448 | 0.5 |
| 13 | 136-143 | YGLDESDL | 911.3996 | 911.3993 | 0.3 |
| 14 | 144-151 | DKVFHLPT | 956.5211 | 956.5200 | 1.1 |
| 15 | 153-161 | TFIGGQESA | 909.4305 | 909.4312 | -0.8 |
| 16 | 163-187 | PLREIIRRLEMAYCQHIGVEFMFIN | 3078.5865 | 3078.5838 | 0.9 |
| 17 | 186-207 | INDLEQCQWIRQKFETPGIMQF | 2724.3279 | 2724.3273 | 0.2 |
| 18 | 207-216 | FTNEEKRTLL | 1250.6742 | 1250.6739 | 0.2 |
| 19 | 220-228 | VRSTRFEEF | 1170.5917 | 1170.5902 | 1.3 |
| 20 | 229-245 | LQRKWSSEKRFGLEGCE | 2053.0306 | 2053.0284 | 1.1 |
| 21 | 246-264 | VLIPALKTIIDKSSENGVD | 2012.1267 | 2012.1274 | -0.3 |
| 22 | 265-291 | YVIMGMPHRGRLNVLANVIRKELEQIF | 3196.7582 | 3196.7598 | -0.5 |
| 23 | 292-326 | CQFDSKLEAADEGSGDVKYHLGMYHRRINRVTDRN | 4080.9581 | 4080.9515 | 1.6 |
| 24 | 327-331 | ITLSL | 546.3501 | 546.3497 | 0.7 |
| 25 | 332-338 | VANPSHL | 737.3941 | 737.3941 | 0.0 |
| 26 | 339-354 | EAADPVVMGKTKAEQF | 1720.8570 | 1720.8574 | -0.3 |
| 27 | 355-375 | YCGDTEGKKVMSILLHGDAAF | 2255.0829 | 2255.0835 | -0.3 |
| 28 | 376-385 | AGQGIVYETF | 1084.5312 | 1084.5310 | 0.3 |
| 29 | 386-406 | HLSDLPSYTTHGTVHVVVNNQ | 2318.1550 | 2318.1524 | 1.1 |
| 30 | 407-434 | IGFTTDPRMARSSPYPTDVARVVNAPIF | 3078.5865 | 3078.5830 | 1.2 |
| 31 | 435-445 | HVNSDDPEAVM | 1213.5153 | 1213.5154 | -0.1 |
| 32 | 446-453 | YVCKVAAE | 882.4385 | 882.4390 | -0.5 |
| 33 | 454-466 | WRSTFHKDVVVDL | 1601.8450 | 1601.8435 | 1.0 |
| 34 | 467-481 | VCYRRNGHNEMDEPM | 1850.7719 | 1850.7731 | -0.7 |
| 35 | 482-487 | FTQPLM | 736.3700 | 736.3698 | 0.2 |

| | | | | | |
|---|---|---|---|---|---|
| 36 | 488-504 | YKQIRKQKPVLQKYAEL | 2133.2548 | 2133.2543 | 0.2 |
| 37 | 505-528 | LVSQGVVNQPEYEEEISKYDKICE | 2799.3407 | 2799.3393 | 0.5 |
| 38 | 529-565 | EAFARSKDEKILHIKHWLDSPWPGFFTLDGQPRSMSC | 4330.1352 | 4330.1325 | 0.6 |
| 39 | 566-573 | PSTGLTED | 819.3732 | 819.3731 | 0.2 |
| 40 | 574-589 | ILTHIGNVASSVPVEN | 1649.8873 | 1649.8857 | 1.0 |
| 41 | 590-596 | FTIHGGL | 744.4050 | 744.4039 | 1.4 |
| 42 | 597-616 | SRILKTRGEMVKNRTVDWAL | 2373.3207 | 2373.3183 | 1.0 |
| 43 | 620-625 | MAFGSL | 625.3010 | 625.3014 | -0.7 |
| 44 | 626-638 | LKEGIHIRLSGQD | 1465.8143 | 1465.8122 | 1.5 |
| 45 | 639-678 | VERGTFSHRHHVLHDQNVDKRTCIPMNHLWPNQAPYTVCN | 4750.2876 | 4750.2837 | 0.8 |
| 46 | 682-687 | SEYGVL | 667.3298 | 667.3297 | 0.1 |
| 47 | 688-693 | GFELGF | 669.3250 | 669.3243 | 1.1 |
| 48 | 694-701 | AMASPNAL | 774.3812 | 774.3815 | -0.3 |
| 49 | 702-708 | VLWEAQF | 892.4561 | 892.4563 | -0.3 |
| 50 | 715-734 | AQCIIDQFICPGQAKWVRQN | 2318.1550 | 2318.1533 | 0.8 |
| 51 | 736-759 | IVLLLPHGMEGMGPEHSSARPERF | 2660.3449 | 2660.3436 | 0.5 |
| 52 | 760-770 | LQMCNDDPDVL | 1262.5412 | 1262.5392 | 1.6 |
| 53 | 771-778 | PDLKEANF | 933.4684 | 933.4676 | 0.8 |
| 54 | 779-783 | DINQL | 602.3142 | 602.3144 | -0.3 |
| 55 | 784-788 | YDCNW | 700.2408 | 700.2395 | 1.8 |
| 56 | 789-833 | VVVNCSTPGNFFHVLRRQILLPFRKPLIIFTPKSLLRHPEARSSF | 5230.9478 | 5230.9455 | 0.4 |
| 57 | 834-842 | DEMLPGTHF | 1046.4593 | 1046.4612 | -1.7 |
| 58 | 843-864 | QRVIPEDGPAAQNPENVKRLLF | 2491.3439 | 2491.3416 | 0.9 |
| 59 | 865-887 | CTGKVYYDLTRERKARDMVGQVA | 2659.3447 | 2659.3443 | 0.1 |
| 60 | 888-902 | ITRIEQLSPFPFDLL | 1788.9894 | 1788.9894 | 0.0 |
| 61 | 903-914 | LKEVQKYPNAEL | 1431.7835 | 1431.7842 | -0.5 |
| 62 | 915-928 | AWCQEEHKNQGYYD | 1770.7188 | 1770.7177 | 0.7 |
| 63 | 929-945 | YVKPRLRTTISRAKPVW | 2071.2293 | 2071.2287 | 0.3 |
| 64 | 946-967 | YAGRDPAAAPATGNKKTHLTEL | 2282.1902 | 2282.1888 | 0.6 |
| 65 | 968-975 | QRLLDTAF | 963.5263 | 963.5258 | 0.6 |
| 66 | 976-992 | DLDVFKNFSLEHHHHHH | 2149.0136 | 2149.0111 | 1.2 |

Table S2.Peptic peptides derived from E2o-h.

| | | Monoisotopic Mass $[M+H]^+$ (Da) | | Error (ppm) |
|---|---|---|---|---|
| **Position** | **Sequence** | **Experimental** | **Theoretical** | |
| 6-33 | VKTPAFAESVTEGDVRWEKAVGDTVAE | 2891.4419 | 2891.4 | -0.1 |
| 21-48 | RWEKAVGDTVAEDEVVCEIETDKTSVQV | 3135.5089 | 3135.5 | -2.0 |
| 51-59 | PANGVIEAL | 883.4881 | 883.5 | -0.3 |
| 59-75 | LLVPDGGKVEGGTPLFT | 1699.9258 | 1699.9 | -0.4 |
| 76-94 | LRKTGAAPAKAKPAEAPAA | 1819.0558 | 1819.1 | 0.5 |
| 95-105 | AAPKAEPTAAA | 997.5299 | 997.5 | -1.4 |
| 106-134 | VPPPAAPIPTQMPPVPSPSQPPSGKPVSA | 2828.5017 | 2828.5 | 0.1 |
| 135-180 | VKPTVAPPLAEAGAGKGLRSEHREKMNRMRQRIAQRLKEAQNTCAM | 5128.7045 | 5128.7 | -0.9 |
| 181-186 | LTTFNE | 724.3506 | 724.4 | -0.8 |
| 187-216 | IDMSNIQEMRARHKEAFLKKHNLKLGFMSA | 3543.8502 | 3543.8 | 0.1 |
| 217-223 | FVKASAF | 769.4239 | 769.4 | -0.5 |
| 224-233 | ALQEQPVVNA | 1068.5677 | 1068.6 | -0.6 |
| 234-246 | VIDDTTKEVVYRD | 1552.7853 | 1552.8 | 0.0 |
| 247-252 | YIDISV | 709.3771 | 709.4 | 0.6 |
| 253-272 | AVATPRGLVVPVIRNVEAMN | 2106.1837 | 2106.2 | -0.7 |
| 273-292 | FADIERTITELGEKARKNEL | 2333.2448 | 2333.2 | -0.5 |
| 293-301 | AIEDMDGGT | 908.3661 | 908.4 | -0.6 |
| 302-313 | FTISNGGVFGSL | 1198.6095 | 1198.6 | -0.7 |
| 314-350 | FGTPIINPPQSAILGMHGIFDRPVAIGGKVEVRPMMY | 4009.0976 | 4009.1 | -0.9 |
| 350-364 | YVALTYDHRLIDGRE | 1820.9286 | 1820.9 | -0.2 |
| 365-368 | AVTF | 437.2396 | 437.2 | 0.3 |
| 369-383 | LRKIKAAVEDPRVLL | 1721.0796 | 1721.1 | 0.0 |

[a] For peptic digestion, samples of E1o-h (25 μL) and E2o-h (25 μl) were prepared in 20 mM $NH_4NCO_3$ (pH 7.0) . Peptic digestion was as reported by us recently ( SI ref S1 and ref S2)

## SI References


S1. Chandrasekhar, K., Wang, J., Arjunan, p., Sax, M., Park, Y. H., Nemeria, N. S., Kumaran, S., Song, J., Jordan, F., and Furey, W. (2013) Insight to the interaction of the dihydrolipoamide acetyltransferase (E2) core with the peripheral components in the *Escherichia coli* pyruvate dehydrogenase complex via multifaceted approaches. *J. Biol. Chem*. **288**, 15402-15417

S2. Wang, J., Nemeria, N. S., Chandrasekhar, K., Kumaran, S., Arjunan, P., Reynolds, SD., Calero, G., Brukh, R., Kakalis, L., Furey, W., and Jordan, F. (2014) Structure and function of the catalytic domain of the dihydrolipoyl acetyltransferase component in *Escherichia coli* pyruvate dehydrogenase complex. *J. Biol. Chem***. 289**, 15215-15230