

Supplementary Material

Critical roles of CTP synthase N-terminal in cytoophidium assembly

Yong Huang^{a,b}, Jin-Jun Wang^b, Sanjay Ghosh^{a,1,*} and Ji-Long Liu^{a,c,*}

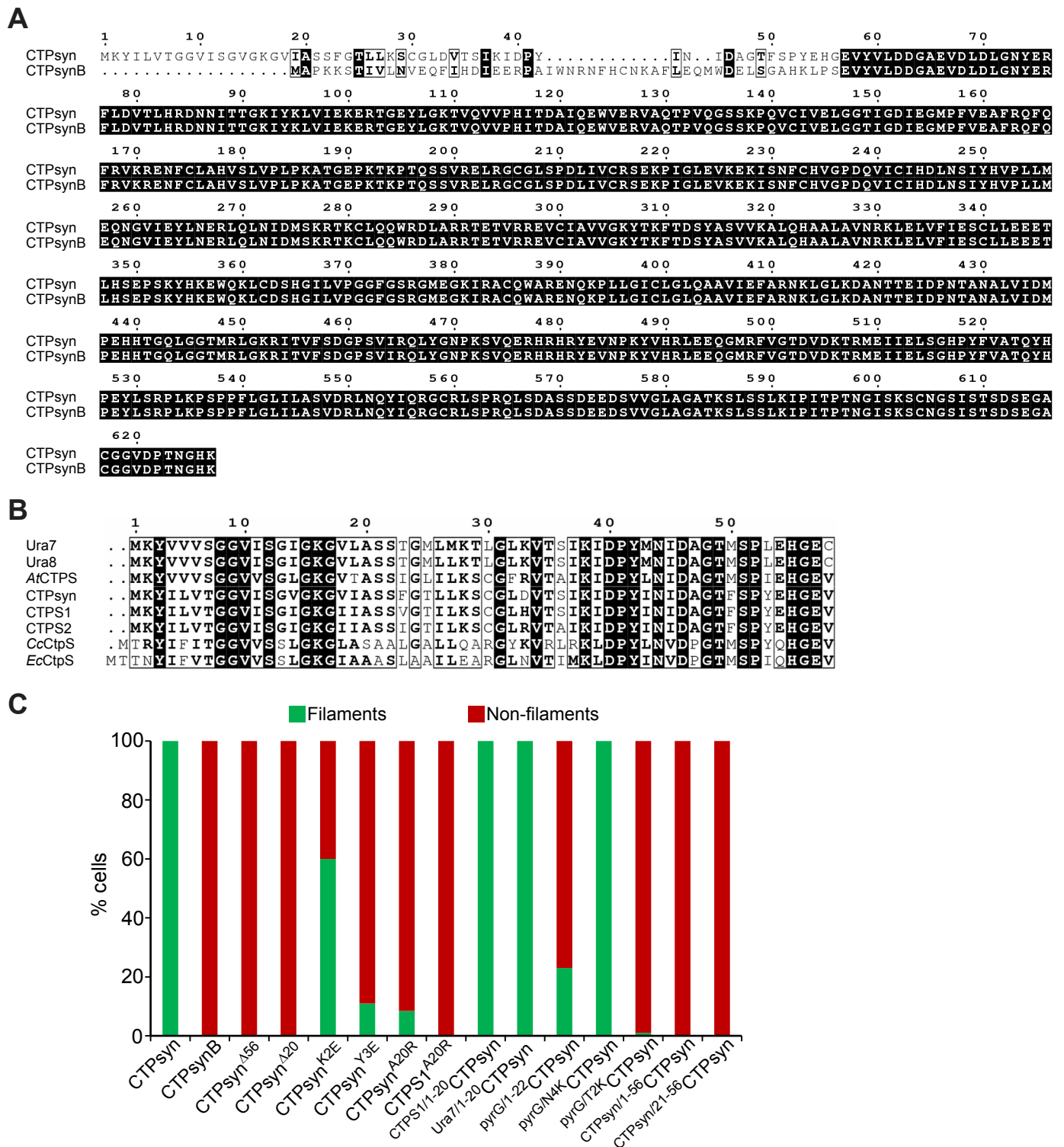


Figure S1. *Drosophila* CTPsyn has a unique N-terminal sequence that is conserved in the orthologous proteins. (A) Amino acid sequence alignment of *Drosophila* CTPsyn and CTPsynB polypeptides using the Clustal omega programme (<http://www.ebi.ac.uk/Tools/msa/clustalo/>) reveals a unique N-terminal for both proteins (56 amino acids for CTPsyn, 52 amino acids for CTPsynB). (B) Multiple sequence alignment of CTPsynthetase N-terminal 56 amino acids from *Drosophila melanogaster* (CTPsyn) with the corresponding CTP synthetase region from *Saccharomyces cerevisiae* (Ura7 and Ura8), *Arabidopsis thaliana* (AtCTPS), human (CTPS1 and CTPS2), *Caulobacter crescentus* (CcCtps), and *Escherichia coli* (EcCtps) proteins. The sequence alignment was performed using Clustal omega and presented using Espirit3 (<http://esprit3.ibcp.fr>). The amino acid position shown at the top of the sequence is from the N-terminal end of the *Drosophila* protein. The identical amino acids are shaded in black. (C) Quantitation of filament-forming capacity of all proteins derived from the constructs generated in this study. Cells showing filamentous structures and clumps/aggregates (non-filaments) were counted and the percentages shown in green and red, respectively, on the y-axis of the stacked barplot. The constructs are shown on the x-axis. At least 75 Venus-positive cells were counted for each construct.

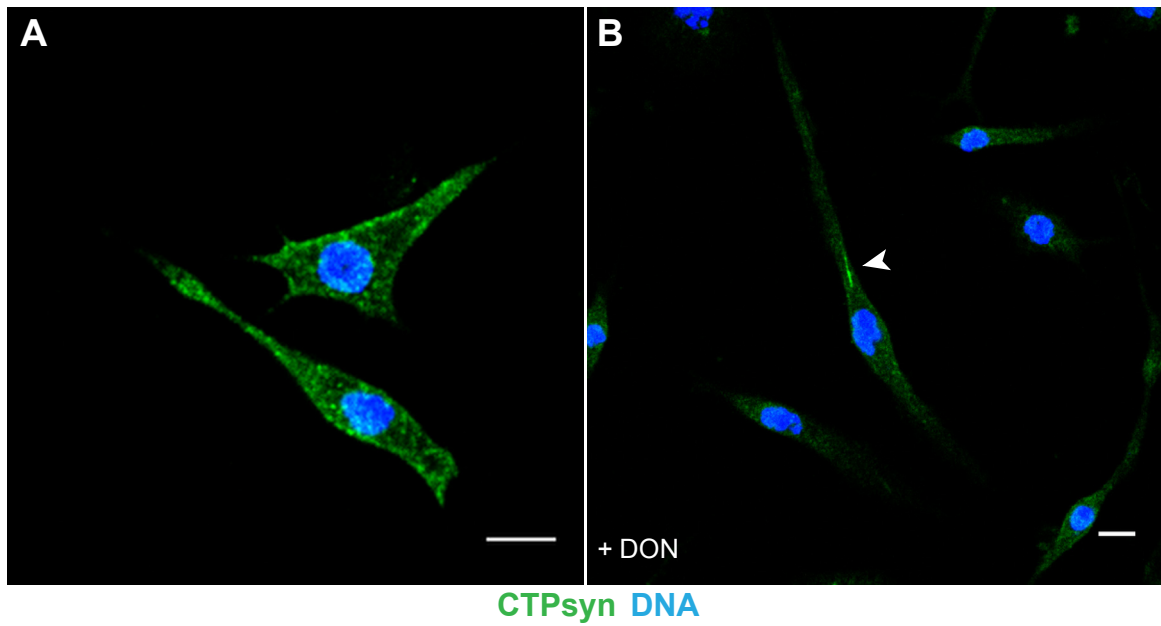


Figure S2. Distribution of CTPsyn in *Drosophila* clone 8 cells.

Immunofluorescence staining of clone 8 cells with anti-CTPsyn antibody in absence (A) or presence of DON (B). The arrowhead shows CTPsyn filament within a cell treated with DON. Hoescht staining of DNA is shown in blue. Scale bar = 5 μm.

Figure S2

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Table S1. List of primers used in this study.

Oligo	Sequences (5' > 3')
OL1	CACCATGAAATACATCCTGGTAACTGGTGG
OL2	CACCATGGCGCCAAAAAAGTCCAC
OL3	CTTATGGCCATTGGTAGGATCAAC
OL4	CACCATGAAGTACATTCTGGTACTGGTG
OL5	GTCATGATTTATTGATGGAACTTC
OL6	CACCATGAAGTACATCCTGGTCACG
OL7	GCTTATTTCCAACCTCAGCTATCC
OL8	GAGGTTTACGTTTTGGACGATGG
OL9	TCCTCGTTCGGAACGCTTTTG
OL10	CATGGTGAAGGGGGCGGC
OL11	ACAAAAAAGCAGGCTCCGCGGCCGCCCCCTTCACCA TGAAGTACATTCTGGTACTGGTGGTGTATATCAGG AATTGGAAAAGGAATCATTGCCTCCTCGTTCGGAAC GCTTTTGAAATCCTGTGGTCT
OL12	ACAAAAAAGCAGGCTCCGCGGCCGCCCCCTTCACCA TGAAGTACGTTGTTGTTTCAGGTGGTGTCAATTCGGG TATCGGTAAAGGTGTTCTTGCATCCTCGTTCGGAACG CTTTTGAAATCCTGTGGTCT
OL13	ACAAAAAAGCAGGCTCCGCGGCCGCCCCCTTCACCA TGACAACGAACATATTTTTGTGACCGGCGGGGTCGT ATCCTCTCTGGGTAAAGGCATTGCCGCATCCTCGTTC GGAACGCTTTTGAAATCCTGTGGTCT
OL14	GCGCCAAAAAAGTCCACCATTG
OL15	CTTTGTACAAAAAAGCAGGCTCCGCGGCCGCCCCCTT CACCATGAAATACATCCTGGTAACTGGTGGCGTCATT AGTGGCGTGGGAAAAGGAGTGATTGCCTCCTCGTTC GGAACGCTTTTGAAATCCTGTGGTCTGGATGTAACCT CGATCAAGATTGACCCCTATATCAATATAGATGCTGG AACCTTTTCGCCTTATGAGCATGGCGGCCAAAAAA GTCCACCATTGTGCTCAATGTGGAGCAGT
OL16	CTTTGTACAAAAAAGCAGGCTCCGCGGCCGCCCCCTT CACCATGTCCTCGTTCGGAACGCTTTTGAAATCCTGT GGTCTGGATGTAACCTCGATCAAGATTGACCCCTATA TCAATATAGATGCTGGAACCTTTTCGCCTTATGAGCA TGGCGGCCAAAAAAGTCCACCATTGTGCTCAATGT GGAGCAGT
OL17	GCCCCCTTCACCATGGAATACATCCTGGTAAC
OL18	GTTACCAGGATGTATTCCATGGTGAAGGGGGC
OL19	CCCCCTTCACCATGAAAGAGATCCTGGTAACTGGTG
OL20	CACCAGTTACCAGGATCTCTTTCATGGTGAAGGGGG
OL21	CTTCACCATGAAATACGCCCTGGTAACTGGTGG
OL22	CCACCAGTTACCAGGGCGTATTTTCATGGTGAAG
OL23	CATGAAATACATCGCGGTAACCTGGTGGC

OL24	GCCACCAGTTACCGCGATGTATTTTCATG
OL25	GAAATACATCCTGGCAACTGGTGGCGTC
OL26	GACGCCACCAGTTGCCAGGATGTATTTTC
OL27	GAAATACATCCTGGTAGCTGGTGGCGTCATTAG
OL28	CTAATGACGCCACCAGCTACCAGGATGTATTTTC
OL29	CATCCTGGTAACTGCTGGCGTCATTAGTG
OL30	CACTAATGACGCCAGCAGTTACCAGGATG
OL31	CTGGTAACTGGTGGCGTCATTAGTGGC
OL32	GCCACTAATGACGGCACCAGTTACCAG
OL33	GTAACCTGGTGGCGCCATTAGTGGCGTG
OL34	CACGCCACTAATGGCGCCACCAGTTAC
OL35	GGTAACTGGTGGCGTCCGTAGTGGCGTGGGAAAAG
OL36	CTTTTCCCACGCCACTACGGACGCCACCAGTTACC
OL37	CTGGTGGCGTCATTAAAGGCGTGGGAAAAGG
OL38	CCTTTTCCCACGCCTTTAATGACGCCACCAG
OL39	GTGGCGTCATTAGTAGCGTGGGAAAAGG
OL40	CCTTTTCCCACGCTACTAATGACGCCAC
OL41	GTCATTAGTGGCGCGGGAAAAGGAGTG
OL42	CACTCCTTTTCCCAGCCACTAATGAC
OL43	CATTAGTGGCGTGGCAAAAAGGAGTGATTGC
OL44	GCAATCACTCCTTTTGGCACGCCACTAATG
OL45	CATTAGTGGCGTGGGAGCAGGAGTGATTGCCTC
OL46	GAGGCAATCACTCCTGCTCCCACGCCACTAATG
OL47	GCGTGGGAAAAGCAGTGATTGCCTCC
OL48	GGAGGCAATCACTGCTTTTCCCACGC
OL49	GTGGCGTGGGAAAAGGACGGATTGCCTCCTCGTTC
OL50	GAACGAGGAGGCAATCCGTCCTTTTCCCACGCCAC
OL51	GAAAAGGAGTGATTTCGCTCCTCGTTCGGAAC
OL52	GTTCCGAACGAGGAGCGAATCACTCCTTTTC
OL53	GAAAAGGAATCATTTCGACAGTGTGGGCAC
OL54	GTGCCACACTGCTGCGAATGATTTCCTTTTC
OL55	GCCCCCTTCACCATGAAAACGAACTATATTTTTG
OL56	CAAAAATATAGTTCGTTTTTCATGGTGAAGGGGGC
OL57	CTTCACCATGACAACGAAATATATTTTTGTGACCGGC
OL58	GCCGGTCACAAAATATATTTTCGTTGTCATGGTGAAG