

doi: 10.1387/ijdb.120256hk

SUPPLEMENTARY MATERIAL

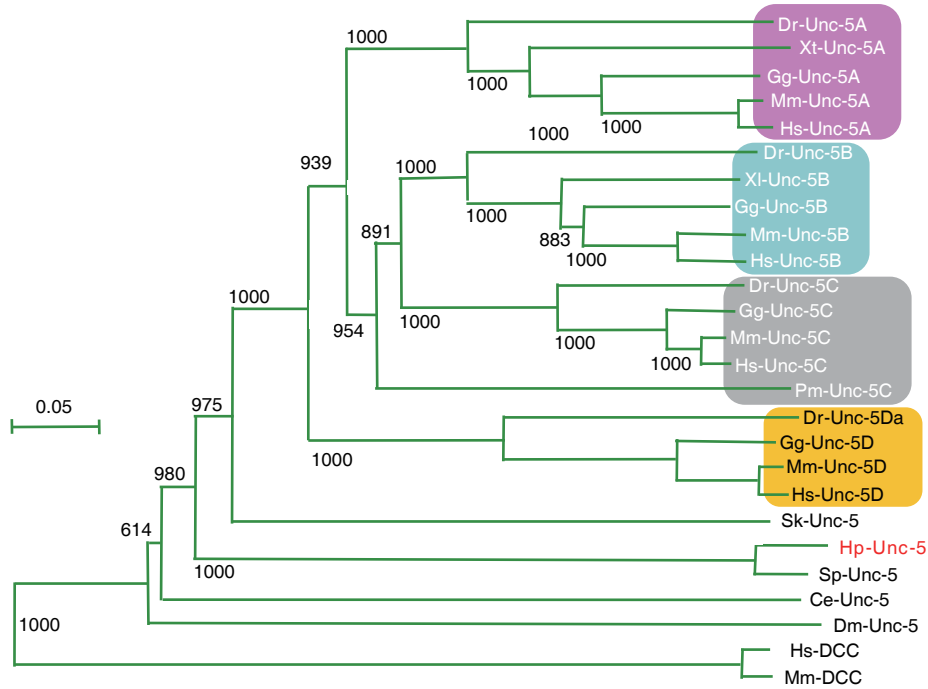
corresponding to:

**Unc-5/netrin-mediated axonal projection
during larval serotonergic nervous system formation
in the sea urchin, *Hemicentrotus pulcherrimus***

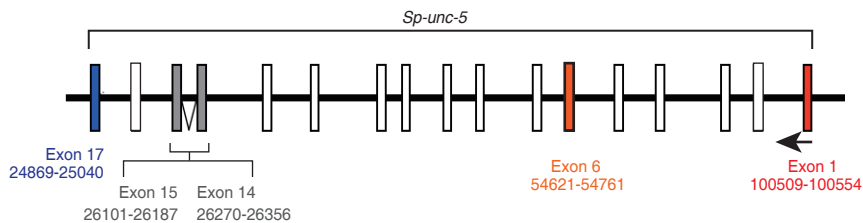
KOUKI ABE, TOMOKO KATOW, SHIOH OOKA and HIDEKI KATOW

*Address correspondence to: Hideki Katow, Research Center for Marine Biology, Tohoku University, Asamushi, Aomori, Aomori 039-3501, Japan.
E-mail: hkatow@m.tohoku.ac.jp

Full text for this paper is available at: <http://dx.doi.org/10.1387/ijdb.120256hk>



Supplementary Fig. S1. Phylogenetic analysis. Phylogenetic tree of *Unc-5* family by neighbor-joining method based on amino acid sequences. Whole protein sequences of Deleted Colorectal Cancer (DCC) of mouse (Mm-DCC; NP_031857) and human (Hs-DCC; NP_005206) were used as outgroups. *Sk-Unc-5*, *Saccoglossus kowalevskii* (NP_001161598); *Ce-Unc-5*, *Caenorhabditis elegans* (NP_500823); *Dm-Unc-5*, *Drosophila melanogaster* (NP_611033); *Dr-Unc-5Da* (XP_003200904), *D. rerio*; *Dr-Unc-5A* (NP_001107937), *Dr-Unc-5B* (NP_001104619), *Dr-Unc-5C* (NP_001093454), *Danio rerio*; *Gg-Unc-5A* (XP_424067), *Gg-Unc-5B* (NP_001026396), *Gg-Unc-5C* (NP_989782), *Gg-Unc-5D* (XP_424529), *G. gallus*; *Hs-Unc-5A* (NP_588610), *Hs-Unc-5B* (NP_734465), *Hs-Unc-5C* (NP_003719), *Hs-Unc-5D* (NP_543148), human; *Mm-Unc-5A* (NP_694771), *Mm-Unc-5B* (NP_084046), *Mm-Unc-5C* (NP_033498), *Mm-Unc-5D* (NP_694775), mouse; *Xt-Unc-5A* (NP_001093674), *X. laevis*; *XI-Unc-5B* (NP_001082302), *Xenopus (Silurana) tropicalis*; *Pm-Unc-5C* (AAU94359), *Petromyzon marinus* (*sea lamprey*); *Sp-Unc-5* (XP_003729771), *S. purpuratus*; *Hp-Unc-5v1* (AB751505), *H. pulcherimus*.



Supplementary Fig. S2. Seventeen-exon structure of *Sp-unc-5C-like* (XM_003729723).

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HpV2          TGC GGCCATTGGTTGGAGCGAATGGAGCGTGTGGTCCCGCATGCACGGACGAGTGTGAAC 60
SpEx6-Int    TGCAGCCATCGGTTGGAGCGAATGGAGCGTGTGGTCTCGC-TGCACGGACGAGTGTGTAC 59
*** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** **
HpV2          AGATCAGGACCAGGACCTGCACCAGGCAATGTTCCGGGCGCATCGCAGCAACGGAGGGACT 120
SpEx6-Int    AGATCAGAACCCAGGACCTGCACCAGGCAATGTTCCGGGCGAACCCGACGCAACGGAGGAACT 119
*** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** **
HpV2          GCTCTGGAGGACTCTGTTAAGTGAGCCACCACGTATGGTGTCTCCCTCTATACTTCTTT 180
SpEx6-Int    GCTCTGGAGGACTCTGTTAAGTGAGCCACCACGTATG-TGTCTCCCTCTATACCAATTT 178
*** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** **
HpV2          TGTTTGAAGGCC---ATGTACAGTAATTGCGAGATTGCAGCGCTGGGAAA----- 229
SpEx6-Int    CATTGAAGGCCATATGTACAGTAATGGCGAGTGTGCAGCTCTGGAAAGAACCAAT 238
*** ** ** ** * ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** **
HpV2          TACTGTTTGTCTACTCAATATGCTAAAAATCTATTC--TGAAATCACTAAAAAGGA 287
SpEx6-Int    TACCATTGCTTATACACAATTTG-TAAAAATCTATTTAATAAAATCACTTACAAGTA 297
*** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** ** **
HpV2          TCATTACTAAAAAAAAAAAAAAAAAAAAA 318
SpEx6-Int    TCATTAA-CAAAAAGAAAAACAAGCAAAA- 326
*** ** ** ** * ** ** **

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Supplementary Fig. S3. DNA sequence alignment of *Hp-unc-5v2* (HpV2) and reverse complementary sequence of *Sp-unc-5* (SpEx6-Int). 3' ORF (T^1-G^{198} or $T^{1172}-G^{1372}$ of Fig. 4) and 3' UTR sequences of *Hp-unc-5v2* ($T^{202}-C^{296}$ or $T^{1375}-C^{1467}$ of Fig. 4) were similar to the structure of *Sp-unc-5C*-like exon-6 adjacent to the intron ($T^{987}-C^{1138}$ or T^1-C^{305} of SpEx6-Int). Blue letters, 3' ORF region; Orange, stop codon; Green, 3' UTR region; Red, poly-A region.

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v1      MRRRSAGLR IHEVTVVLVLFGA IASRPVLGQLDGQPPQDVPTTMTPLLSQFFIVPQDSY 60
v2      MRRRSAGLR IHEVTVVLVLFGA IASRPVLGQLDGQPPQDVPTTMTPLLSQFFIVPQDSY 60
*****

v1      ITKRSVDLECKAGSPNVYF I CNDEK IADARTH SRGTFDEYYENIRHIALTITKDEVQEY 120
v2      ITKRSVDLECKAGSPNVYF I CNDEK IADARTH SRGTFDEYYENIRHIALTITKDEVQEY 120
*****

v1      FGDEDFWCVCEAAADPQPIRTEKAYIREAYLRKQFLQMPLDHSVPLHDKFHLLCRAPEGV 180
v2      FGDEDFWCVCEAAADPQPIRTEKAYIREAYLRKQFLQMPLDHSVPLHDKFHLLCRAPEGV 180
*****

v1      PEPTIHWEIDGVPIDDENLVHYVVTYDGLIVNEATLADNGNYTCVATNVATYRTTDPAR 240
v2      PEPTIHWEIDGVPIDDENLVHYVVTYDGLIVNEATLADNGNYTCVATNVATYRTTDPAR 240
*****

v1      VIVYDNTNDGAWTMMTEWSTCTGDCDGGTRRRMRYCTNPAPLSMEPTARERHCRPKTVQS 300
v2      VIVYDNTNDGAWTMMTEWSTCTGDCDGGTRRRMRYCTNPAPLSMEPTARERHCRPKTVQS 300
*****

v1      IAQVRPLVGANGACGPACTDECEQIRTRCTRQCSGASQRRDCSGGLCLSEPLLVD DPG 360
v2      IAQVRPLVGANGACGPACTDECEQIRTRCTRQCSGASQRRDCSGGLCLSEPPRMVSPS 360
***** *

v1      IFSPSAVDTT SQNPAAGSKNGLSKQIPVYIGISLAIVL LLLVFLFIAIYLVTKRKRGNP 420
v2      ILLFEGHVQ----- 370
*

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Supplementary Fig. S4. Protein alignment of Hp-Unc-5v1 (v1) and v2 (v2) variants shows distinctive structure at carboxyl terminus of v2 variant with v1 (red letters). Orange letter, signaling peptide sequence; Pink, immunoglobulin-like domain-1 and 2; Green, thrombospondin type-1 domain-1; Blue, transmembrane domain.