Supplementary materials

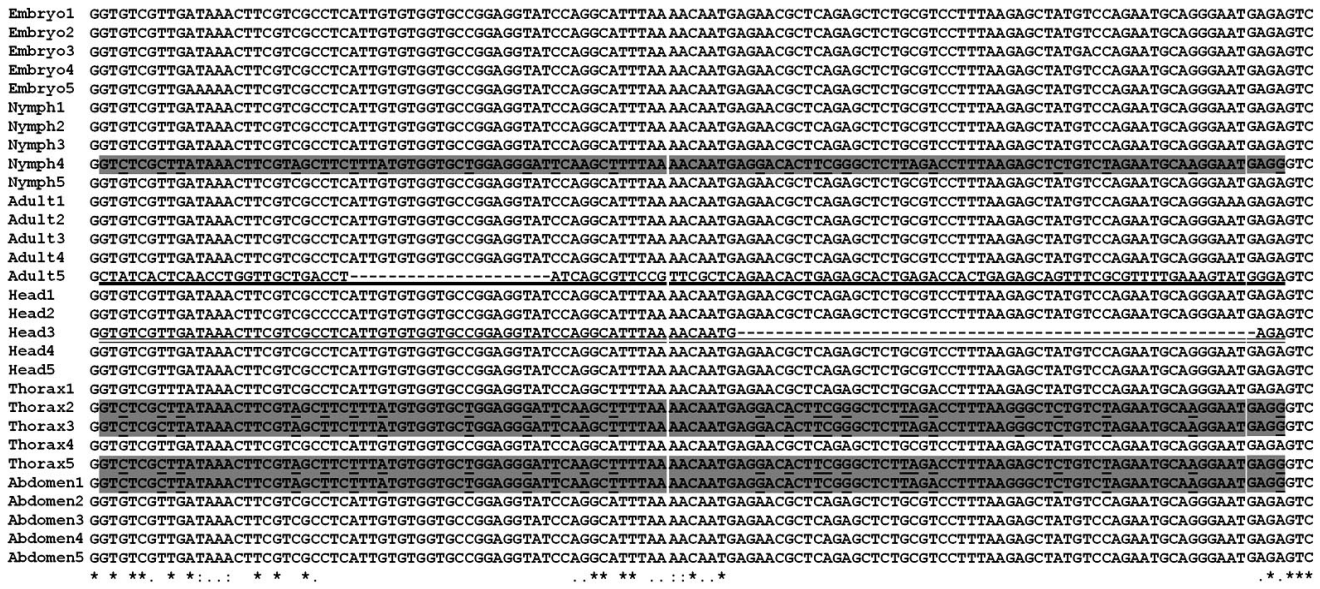

Figure S1. Alignment of 30 clones from different developmental stages and tissues to detect mutually exclusive exon 27A, 27B, 27C and 27D of *LbVGSC*.

Only the region with nucleotides change was shown. 27A was underlined. 27C was double underlined. 27D was highlighted in black and the 25 nucleotides different from 27B were underlined.

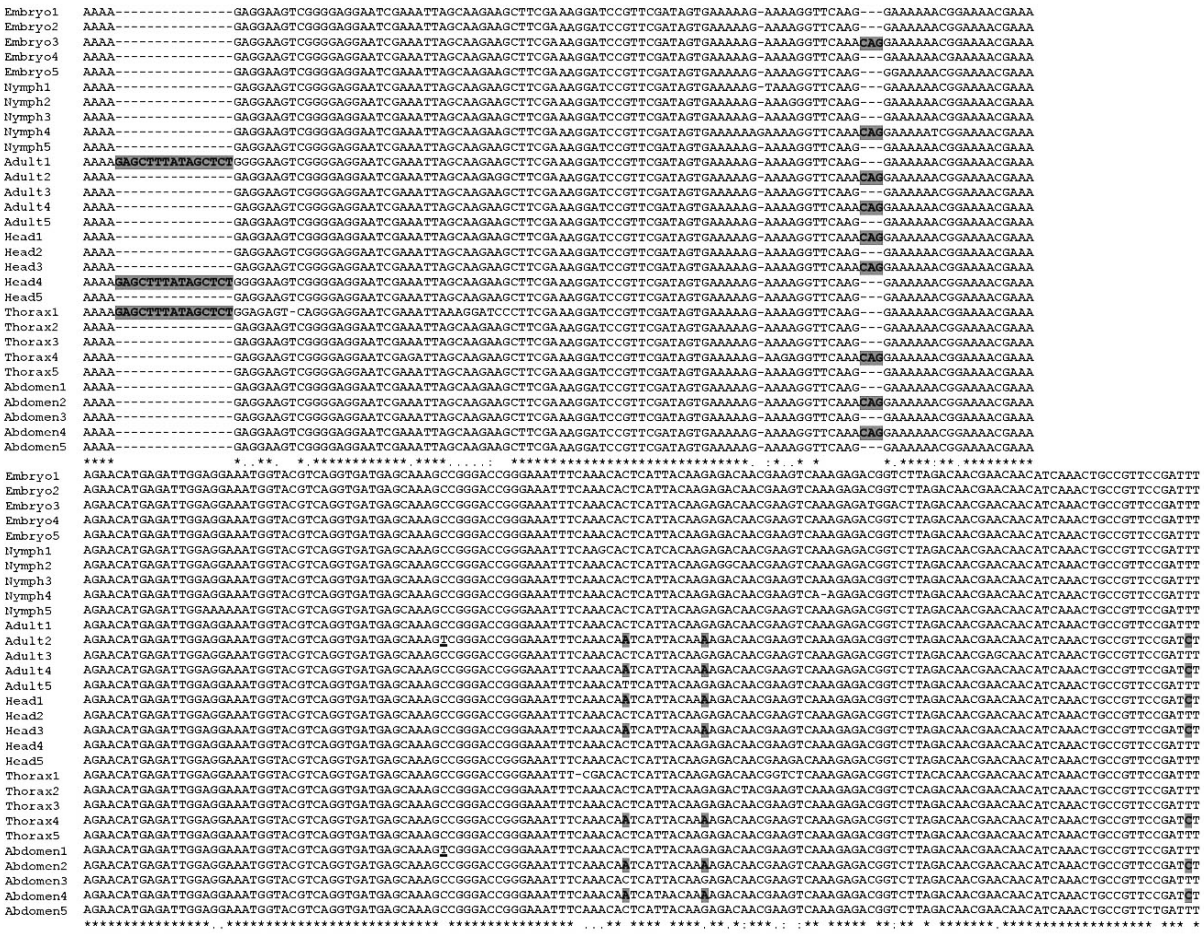

Figure S2. Alignment of 30 clones from different developmental stages and tissues to detect exon 11 of *LbSC1*. Only the region containing nucleotides change was shown. Exon 11 was highlighted in black. The fisrt C to T change was underlined. Other highlighted bases in black were the same with the corresponding genomic DNA sequence.

Table S1. Primers used in cloning the full length cDNAs and in qPCR analysis.

Fragments names	Primer names	Primer sequences (5′–3′)	Length (bp)
Primers for cDNA cloning of *LbVGSC*			
a	a-F (NUP)	AAGCAGTGGTAACAACGCAGAGT	480
	a-R	GTTCCATGCGTCTCTAAGGTAAG	
b	b-F	TCCAACAATAGAATCCACTGAAG	723
	b-R	AACGAGCCAAGGAAGATAATGAC	
c	c-F	TTTCGGACCCAACCCAAACTAC	1782
	c-R	ATTCCCATGACGGCAAAGATG	
d	d-F	CATTGTGGCCTTGTCATTACTC	1709
	d-R	GCRAANARYTGNACNCCCAT	
e	e-F	TCGAAACTGCTGTCATCACTATG	2321
	e-R	TGGACGGGCTCAGACG	
Primers for cDNA cloning of *LbSC1*			
a1	a1-F	AAGTGATGTTTCAAGCCAATCTC	1124
	a1-R	TTCCCAATAGTCTAACGTAATCAAC	
b1	b1-F	GAGAACTCCGAAATAAGTGCGTCT	4451
	b1-R	CGCTAAGTATGCTTGACCAACGTG	
c1	c1-F	ATGGCAGGGAATGAGGATTGTAG	2583
	c1-R	AATTATCAGCTCCCCTGTTCTTC	
Primers for qPCR and RT-PCR			
α-tubulin	α-tubulin-F	AAATCGTTTCCTCGATCACG	259
	α-tubulin-R	ACCATCTGATTGGCAGGTTC	
LbVGSC	LbVGSC-F	GGCTATGTCACTACCCGCTC	211
	LbVGSC-R	AGGCGAACCAATCTCGTTGT	
LbSC1	LbSC1-F	ATCATCAAGGGCTTCGGACG	150
	LbSC1-R	GAGGCTTCGCTGACCGTAAT	

F: forward primer; R: reverse primer.

Table S2. Primers used in cloning genomic DNA containing the identified alternative exons and in RT-PCR analysis.

Amplified regions	Primer names	Primer sequences (5′–3′)
Primers flanking splicing sites to clone genomic DNA containing the alternative exons		
Exon 23	F	GGCGATGGGATGGAATTTACAATAC
	R	ATCTTCGTCGTGGTGTGTCAAGTAG
Exon	F	TCTCAACCTCTTCTTGGCTTTGT
27A/27B/27C/27D	R	TTTCGATTCCTCCCCGACTT
Exon 11	F	AGCTTTGGCCTTGGAAGATGTTCAC
	R	CGTTGAAGATAGATGGTATGGCTTG
Region B	F	CCGACCAAAGAGACAACGGA
	R	GCTCGTCCATTTCTGCCCTA
Primers used in RT-PCR		
Exon 23	23-F	ACTGCCGATAACGACACCAA
	23-R	TCGCCTTCATTCTCGAGGTC
Exon	27-F	AGCTTTGGCCTTGGAAGATGT
27A/27B/27C/27D	27-R	CCGGCAAATAACTGCACTCC
Exon 11	11-F	TCTCAACCTCTTCTTGGCTTTGT
	11-R	ATATTGTCGGGGGATGGAGC
Exon 13A/13B	13A/13B-F	CAAAGACTCAGCGACGGAGA
	13A/13B-R	GCTCGTCCATTTCTGCCCTA
	13A/13B -R1	AACAGGAAAGGCTTTGATTTG

F: forward primer; R: reverse primer.

Table S3. Voltage-gated sodium channels used in phylogenetic analysis.

Species	GenBank accession numbers	Length (bp)	pI	Molecular weight (kDa)
Voltage-gated sodium channels				
Bombus terrestris	XP_003397764	2044	5.22	232.1
Nasonia vitripennis	CAM31896	2084	5.10	235.6
Apis mellifera	ACV87000	2044	5.22	232.2
Camponotus floridanus	EFN61422	2088	5.05	237.5
Harpegnathos saltator	EFN86793	2055	5.30	234.0
Blattella germanica	AAC47484	2031	5.14	229.9
Periplaneta americana	ACX44801	2050	5.20	232.0
Tribolium castaneum	ACV87003	2048	5.34	231.9
Cimex lectularius	ACI43362	2027	**5.41**	229.1
Pediculus humanus	AAP20107	2051	5.08	233.2
*Liposcelis bostrychophila**	KC699919	2014	5.12	232.8
Culex quinquefasciatus	BAI77917	2149	5.13	240.5
Aedes aegypti	ACB37023	2140	5.06	240.2
Culex pipiens pallens	BAI77918	2147	5.13	240.4
Anopheles gambiae	CAM12801	2139	5.07	240.0
*Aedes albopictus**	AAT69680	2058	5.07	230.6
Musca domestic	AAB47604	2105	**4.95**	236.4
Drosophila melanogaster	NP_001188635	2143	5.04	240.6
Drosophila virilis	XP_002055012	2171	5.06	244.2
Drosophila grimshawi	XP_001992511	2171	5.08	243.8
Drosophila willistoni	XP_002071221	**2201**	5.11	**247.1**
Drosophila mojavensis	XP_002010837	2168	5.08	243.1
Drosophila erecta	XP_001977935	2130	5.02	239.3
Drosophila ananassae	XP_001966146	2136	5.03	239.5
Bactrocera dorsalis	JN416983	2134	5.16	240.7
Helicoverpa zea	ADF80418	**1830**	5.07	**207.5**
*Plutella xylostella**	BAF37094	1890	4.99	215.4
Bombyx mandarina	ACD80428	1840	4.98	209.2
Bombyx mori	ACJ09096	2038	5.09	231.0

Those indicated with asterisks were partial. The maximum and minimum of length, pI and molecular weight were bolded and underlined.

Table S4. DSC1 orthologs used in phylogenetic analysis.

Species	GenBank accession number	Length (bp)	pI	Molecular weight (kDa)
DSC1 channels				
Apis mellifera	XM_395121	2525	6.56	287.1
Bombus impatiens	XM_003493288	2532	6.57	287.0
Megachile rotundata	XM_003704464	2439	7.35	277.1
Apis florea	XM_003697572	2454	6.46	279.4
Harpegnathos saltator	EFN89067	1966	7.22	223.9
Camponotus floridanus	EFN62327	1975	7.64	225.5
Acromyrmex echinatior	EGI69876	2004	8.03	229.2
Anopheles gambiae	XM_308670	**2961**	6.56	**333.0**
Drosophila melanogaster	NM_001259579	2806	**6.20**	316.5
Drosophila pseudoobscura	XM_002138166	2473	6.69	282.4
Drosophila grimshawi	XM_001987505	2487	6.38	283.5
Drosophila yakuba	XM_002092876	2362	**6.20**	270.0
Drosophila erecta	XM_001976656	2362	6.25	270.0
Drosophila ananassae	XM_001960375	2316	6.26	264.3
Danaus plexippus	EHJ64356	1991	7.28	227.0
Bombyx mori	NM_001258361	2225	7.37	253.4
Nilaparvata lugens	JN619367	2122	6.95	240.6
Acyrthosiphon pisum	XM_001943495	**1896**	6.73	**214.7**
Blattella germanica	AF312365	2304	6.24	260.8
Pediculus humanus	XM_002425122	2203	**8.65**	251.7
Tribolium castaneum	XM_970570	2283	8.06	259.7
Liposcelis bostrychophila	KC699920	2535	8.43	287.2

The maximum and minimum of length, *pI*, and molecular weight were bolded and underlined.

Table S5. Voltage-gated calcium channels used in phylogenetic analysis.

Species names	GenBank accession number	Length (bp)	pI	Molecular weight (kDa)
Voltage-gated calcium channels				
Musca domestica	Q25452	1687	7.25	193.8
Anopheles gambiae	EF595743	1893	6.35	213.6
*Aedes aegypti**	XP_001661548	1815	6.11	206.2
Drosophila melanogaster	AAA81883	2516	**5.17**	276.5
Drosophila simulans	XP_002079606	2537	5.21	278.7
Drosophila virilis	XP_002057533	2563	5.18	283.0
Drosophila grimshawi	XP_001988771	**2732**	5.38	**301.7**
Drosophila sechellia	XP_002038138	2549	5.27	280.3
Drosophila yakuba	XP_002089976	2554	5.31	280.7
*Drosophila erecta**	XP_001969022	2522	5.24	276.9
Pediculus humanus corporis	XM_002431028	**1683**	**7.90**	**192.1**
Bombus terrestris	XP_003393703	1948	6.03	220.3
*Bombus impatiens**	XP_003490120	1916	6.08	217.4
Apis florea	XP_003697046	2285	7.03	255.3
Tribolium castaneum	XP_001807530	1913	6.16	216.5
Acyrthosiphon pisum	XP_003246635	1811	7.46	206.8

Those indicated with asterisks were partial. The maximum and minimum of length, pI and molecular weight were bolded and underlined.