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Supplementary materials

Molecular cloning and gene expression of Type I suppressor of cytokine signaling 6 and 7 (SOCS6 and SOCS7) in white leg shrimps (*Liptopenaeus vannamei*)

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Figure S1: The LvSOCS6 protein coding nucleotide region (top) and its deduced amino acid sequence (bottom). The start (ATG) and stop (TGA) codons are presented in red color. In the amino acid sequence, SH2 and SOCS-box domain positions are shown in red and blue color boxes.

```
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TGTCAGCGGGAACAG TGTCGGGAGGAGTAC CAGAGGAAGGAGAGC AACTGGTGGGCTGTA  180
 C  Q  R  E  Q   C  R  E  E  Y   Q  R  K  E  S   N  W  W  A  V    60
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 G  G  S  K  G   S  R  K  S  S   I  P  L  L  G   L  S  C  G  S    80
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 C  R  G  L  D   D  H  V  F  E   D  L  G  G  V   *               955
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Figure S2: The LvSOCS7 protein coding nucleotide region (top) and its deduced amino acid sequence (bottom). The start (ATG) and stop (TGA) codons are presented in red color. In the amino acid sequence, SH2 and SOCS-box domain positions are shown in red and blue color boxes.

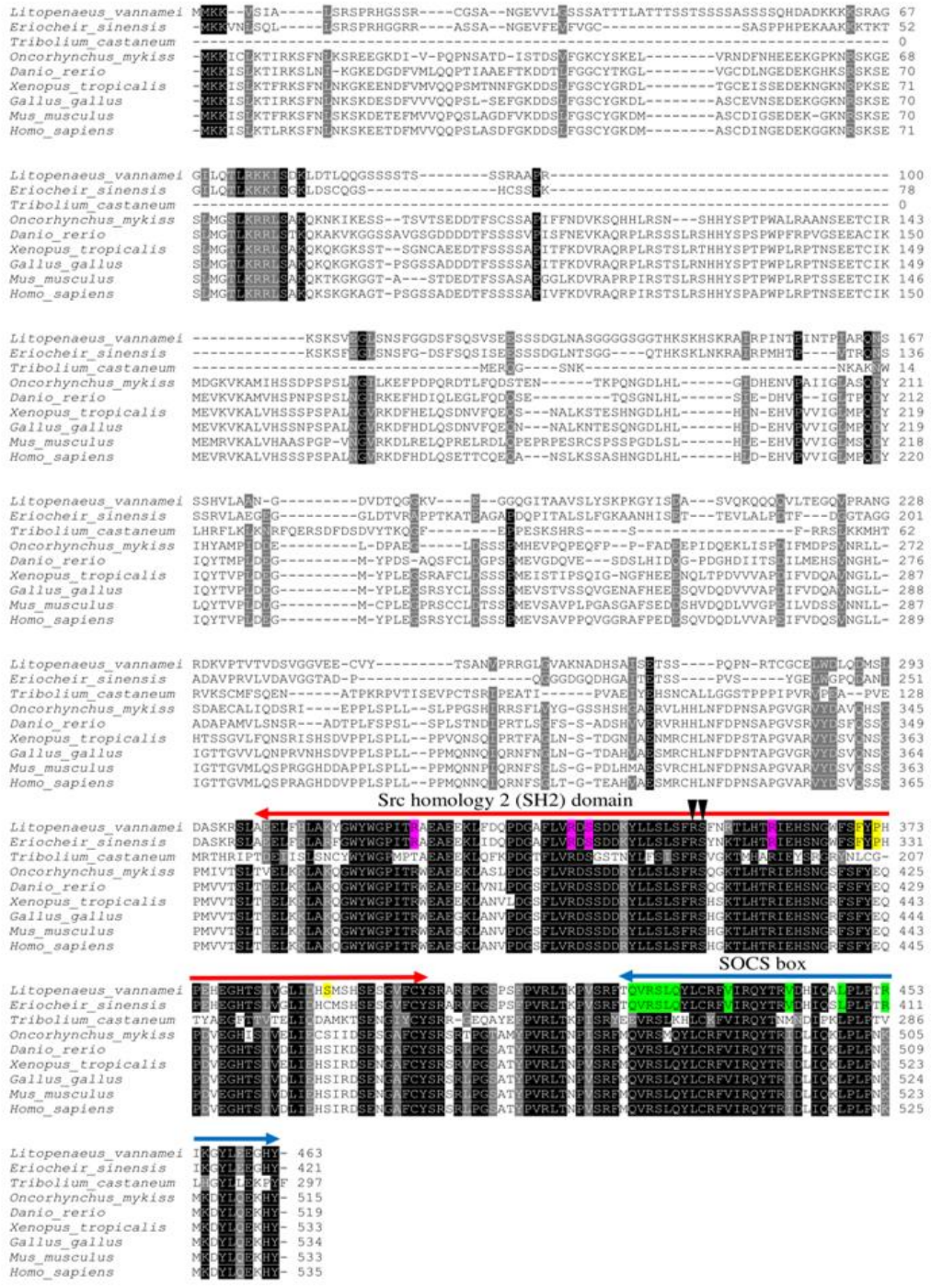
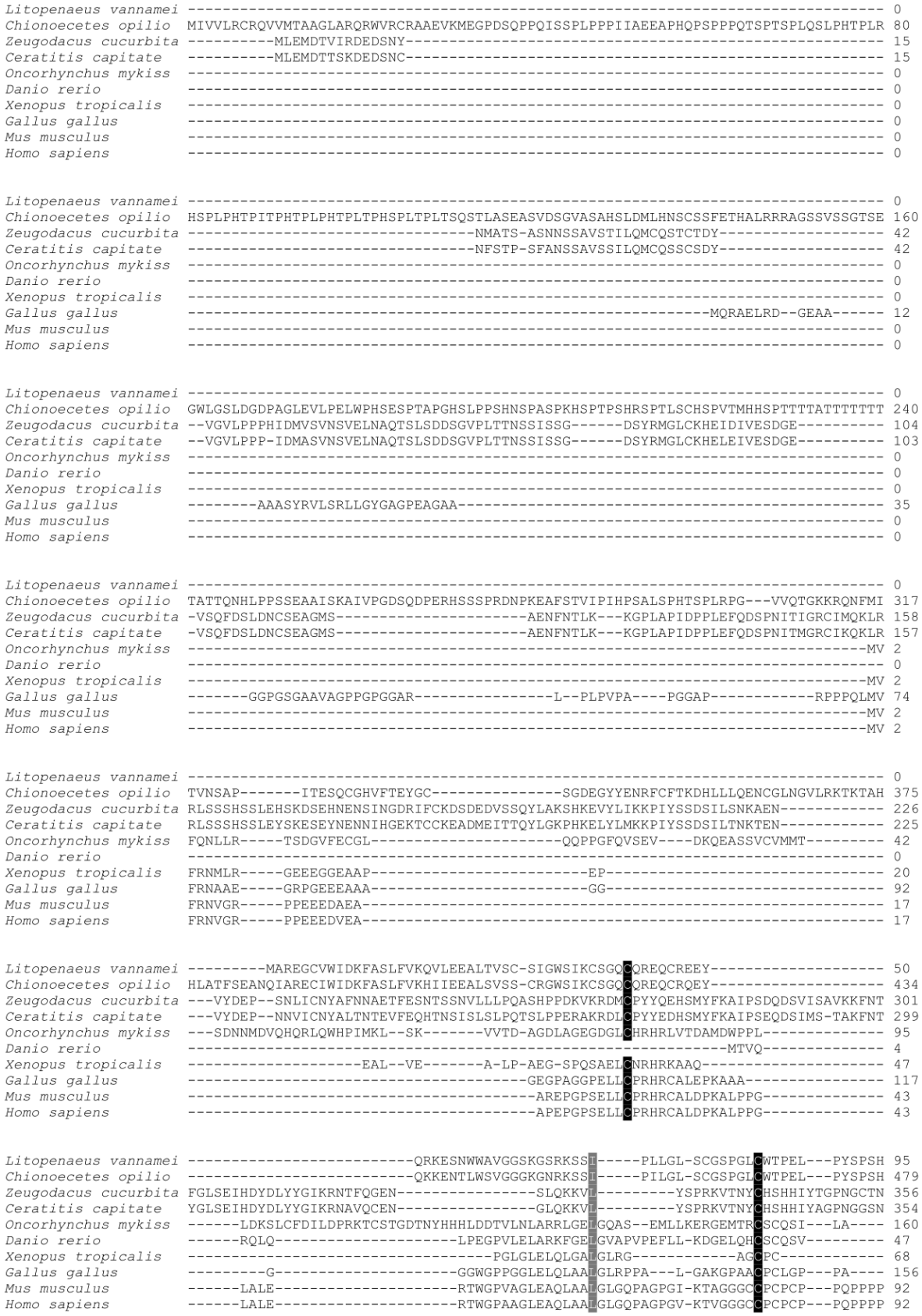


Figure S3: Multiple sequence alignment (MSA) of LvSOCS6 and its counterparts. Strongly conserved and similar residues are highlighted by black and gray shading and the conserved phosphotyrosine recognition site (pY) of LvSOCS6 is indicated with (▼). The regions of the phosphotyrosine binding pocket, hydrophobic binding pocket, and putative elongin B/C binding pockets are indicated in purple, yellow, and green color respectively. The species and the GenBank accession numbers used for the MSA were as follows: SOCS6 from *H. sapiens* (NP_004223.2), *M. musculus* (NP_061291.2), *G. gallus* (NP_001120784.1), *X. tropicalis* (NP_001096240.1) *D. rerio* (XP_687041.2) *O. mykiss* (NP_001182102.1), *E. sinensis* (ATW63847.1) and *T. castaneum* (XP_008190646).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Zeugodacus cucurbita
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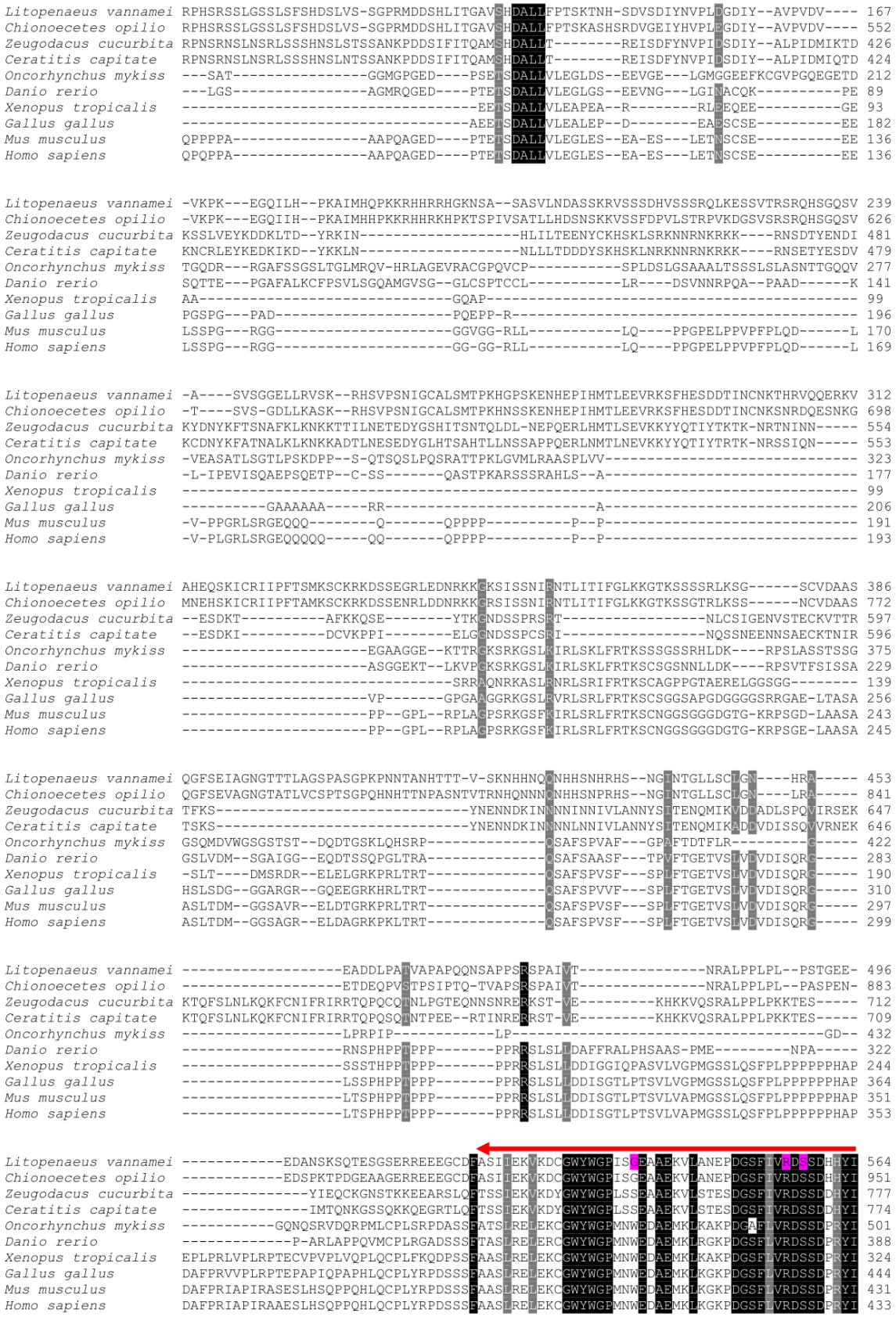
Mus musculus
Homo sapiens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Chionoecetes opilio RPHSRSSLGSSLSFSHDSLVS-SGPRMDDSHLITGAVSHDALLFPTSKASHSRDVGEIYHVPLEGDIY--AVPVDV---- 552
Zeugodacus cucurbita RPNSRNSLNSRLSSSHNSLSTSSANKPDDSIFITQAMSHDALLT--------REISDFYNVPIDSDIY--ALPIDMIKTD 426
Ceratitis capitate RPNSRNSLNSRLSSSHNSLNTSSANKPDDSIFITQAMSHDALLT--------REISDFYNVPIDSDIY--ALPIDMIQTD 424
Oncorhynchus mykiss ---SAT----------------GGMGPGED----PSETSDALLVLEGLDS--EEVGE---LGMGGEEFKCGVPGQEGETD 212
Danio rerio ---LGS----------------AGMRQGED----PTETSDALLVLEGLGS--EEVNG---LGINACQK----------PE 89
Xenopus tropicalis ----------EETSDALLVLEAPEA--R--------RLDEQEE----------GE 93
Gallus gallus ----------AEETSDALLVLEALEP--D--------EAESCSE----------EE 182
Mus musculus QPPPPA----------------AAPQAGED----PTETSDALLVLEGLES--EA-ES---LETNSCSE----------EE 136
Homo sapiens QPQPPA----------------AAPQAGED----PTETSDALLVLEGLES--EA-ES---LETNSCSE----------EE 136
Litopenaeus vannamei -VKPK---EGQILH--PKAIMHQPKKRHHRRHGKNSA--SASVLNDASSKRVSSSDHVSSSRQLKESSVTRSRQHSGQSV 239
Chionoecetes opilio -VKPK---EGQIIH--PKAIMHHPKKRHHRKHPKTSPIVSATLLHDSNSKKVSSFDPVLSTRPVKDGSVSRSRQHSGQSV 626
Zeugodacus cucurbita KSSLVEYKDDKLTD--YRKIN----------HLILTEENYCKHSKLSRKNNRNKRKK----RNSDTYENDI 481
Ceratitis capitate KNCRLEYKEDKIKD--YKKLN----------NLLLTDDDYSKHSKLNRKNNRNKRKK----RNSETYESDV 479
Oncorhynchus mykiss TGQDR---RGAFSSGSLTGLMRQV-HRLAGEVRACGPQVCP----------SPLDSLGSAAALTSSSLSLASNTTGQQV 277
Danio rerio SQTTE---PGAFALKCFPSVLSGQAMGVSG--GLCSPTCCL----------LR----DSVNNRPQA--PAAD------K 141
Xenopus tropicalis AA----------GQAP---------- 99
Gallus gallus PGSPG---PAD----------PQEPP-R---------- 196
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Homo sapiens LSSPG---RGG----------GG-GG-RLL----------LQ----PPGPELPPVPFPLQD------L 169
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Chionoecetes opilio -T----SVS-GDLLKASK--RHSVPSNIGCALSMTPKHNSSKENHEPIHMTLEEVRKSFHESDDTINCNKSNRDQESNKG 698
Zeugodacus cucurbita KYDNYKFTSNAFKLKNKKTTILNETEDYGSHITSNTQLDL-NEPQERLHMTLSEVKKYYQTIYTKTK-NRTNINN----- 554
Ceratitis capitate KCDNYKFATNALKLKNKKADTLNESEDYGLHTSAHTLLNSSAPPQERLNMTLNEVKKYYQTIYTRTK-NRSSIQN----- 553
Oncorhynchus mykiss -VEASATLSGTLPSKDPP--S-QTSQSLPQSRATTPKLGVMLRAASPLVV---------- 323
Danio rerio -L-IPEVISQAEPSQETP--C-SS-------QASTPKARSSSRAHLS--A---------- 177
Xenopus tropicalis ---------- 99
Gallus gallus ----------GAAAAAA-----RR----------A---------- 206
Mus musculus -V-PPGRLSRGEQQQ--------Q-------QPPPP----------P--P---------- 191
Homo sapiens -V-PLGRLSRGEQQQQQ-----QQ-------QPPPP----------P--P---------- 193
Litopenaeus vannamei AHEQSKICRIIPFTSMKSCKRKDSSEGRLEDNRKKGKSISSNIRNTLITIFGLKKGTKSSSSRLKSG------SCVDAAS 386
Chionoecetes opilio MNEHSKICRIIPFTAMKSCKRKDSSENRLDDNRKKGRSISSNIRNTLITIFGLKKGTKSSGTRLKSS------NCVDAAS 772
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Ceratitis capitate --ESDKI----------DCVKPPI--------ELGGNDSSPCSRI----------NQSSNEENNSAECKTNIR 596
Oncorhynchus mykiss ----------EGAAGGE--KTTRGKSRKGSLKIRLSKLFRTKSSSGSSRHLDK----RPSLASSTSSG 375
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Xenopus tropicalis ----------SRRAQNRKASLRNRLSRIFRTKSCAGPPGTAERELGGSGG-------- 139
Gallus gallus ----------VP-------GPGAAGGRKGSLRVRLSRLFRTKSCSGGSAPGDGGGGSRRGAE-LTASA 256
Mus musculus ----------PP--GPL--RPLAGPSRKGSFKIRLSRLFRTKSCNGGSGGGDGTG-KRPSGD-LAASA 243
Homo sapiens ----------PP--GPL--RPLAGPSRKGSFKIRLSRLFRTKSCNGGSGGGDGTG-KRPSGE-LAASA 245
Litopenaeus vannamei QGFSEIAGNGTTTLAGSPASGPKPNNTANHTTT-V-SKNHHNQQNHHSNHRHS--NGINTGLLSCLGN----HRA----- 453
Chionoecetes opilio QGFSEVAGNGTATLVCSPTSGPQHNHTTNPASNTVTRNHQNNNQNHHSNPRHS--NGINTGLLSCLGN----LRA----- 841
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Ceratitis capitate TSKS----------YNENNDKINNNNLNNIVLANNYSITENQMIKADPVDISSQVVRNEK 646
Oncorhynchus mykiss GSQMDVWGSGSTST--DQDTGSKLQHSRP----------QSAFSPVAF---GPAFTDTFLR---------G----- 422
Danio rerio GSLVDM--SGAIGG--EQDTSSQPGLTRA----------QSAFSAASF---TPVFTGETVSLVPVDISQRG----- 283
Xenopus tropicalis -SLT----DMSRDR--ELELGRKPRLTRT----------QSAFSPVSF---SPLFTGETVSLVPVDISQRG----- 190
Gallus gallus HSLSDG--GGARGR--GQEEGRKHRLTRT----------QSAFSPVVF---SPLFTGETVSLVPVDISQRG----- 310
Mus musculus ASLTDM--GGSAVR--ELDTGRKPRLTRT----------QSAFSPVSF---SPLFTGETVSLVPVDISQRG----- 297
Homo sapiens ASLTDM--GGSAGR--ELDAGRKPKLTRT----------QSAFSPVSF---SPLFTGETVSLVPVDISQRG----- 299
Litopenaeus vannamei ----------EADDLPATVAPAPQQNSAPPSRSPAIVT----------NRALPPLPL--PSTGEE- 496
Chionoecetes opilio ----------ETDEQPVSTPSIPTQ-TVAPSRSPAIVT----------NRALPPLPL--PASPEN- 883
Zeugodacus cucurbita KTQFSLNLKQKFCNIFRIRRTQPQCQTNLPGTEQNNSNRERKST-VE---------KHKKVQSRALPPLPKKTES----- 712
Ceratitis capitate KTQFSLNLKQKFCNIFRIRRTQPQSQTNTPEE--RTINRERRST-VE---------KHKKVQSRALPPLPKKTES----- 709
Oncorhynchus mykiss ----------LPRPIP----------LP----------GD-- 432
Danio rerio ----------RNSPHPPTPPP-------PPRRSLSLLDAFFRALPHSAAS-PME----------NPA----- 322
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Litopenaeus vannamei ----------EDANSKSQTESGSERREEEGCDFASILEKVKDCGWYWGPISGEAAEKVLANEPDGSFIVRDSSDHFYI 564
Chionoecetes opilio ----------EDSPKTPDGEAAGERREEEGCDFASILEKVKDCGWYWGPISGEAAEKVLANEPDGSFIVRDSSDHFYI 951
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Ceratitis capitate ----------IMTQNKGSSQKKQEGRTLQFTSSIEKVKDYGWYWGPLSSEAAEKVLSTESDGSFIVRDSSDDFYI 774
Oncorhynchus mykiss ----------GQNQSRVDQRPMLCPLSRPDASSFATSLREEEKCGWYWGPMNWEDAEMKLKAKPDGAFLVRDSSDPRYI 501
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Xenopus tropicalis EPLPRLVPLRPTECVPVPLVQPLQCPLFKQDPSSFAASLREEEKCGWYWGPMNWEDAEMKLKAKPDGSFLVRDSSDPRYI 324
Gallus gallus DAFPRVVPLRPTEPAPIQPAPHLQCPLYRPDSSSFAASLREEEKCGWYWGPMNWEDAEMKLKGKPDGSFLVRDSSDPRYI 444
Mus musculus DAFPRIAPIRASESLHSQPPQHLQCPLYRPDSSSFAASLREEEKCGWYWGPMNWEDAEMKLKGKPDGSFLVRDSSDPRYI 431
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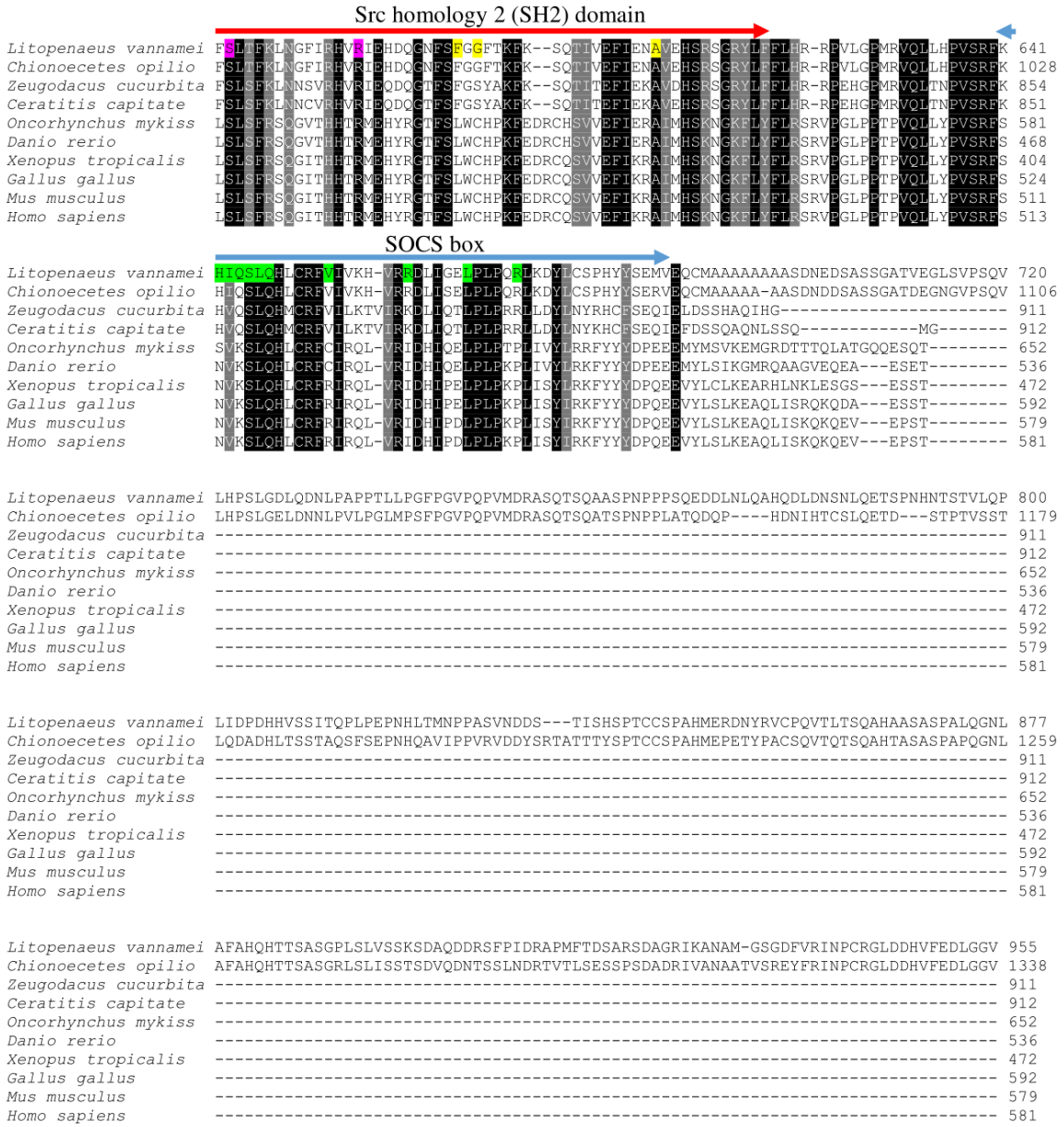

Figure S4: Multiple sequence alignment (MSA) of LvSOCS7 and its counterparts (Only the regions associated with SH2 and SOCS box are represented in the MSA analysis). Strongly conserved and similar residues are highlighted by black and gray shading and the regions of phosphotyrosine binding motif, hydrophobic binding motif, and putative elongin B/C binding motifs are indicated in purple, yellow, and green color respectively. The species and the GenBank accession numbers used for the MSA were as follows: *C. opilio* (KAG0725835.1), *Z. cucurbita* (JAD11819), *C._capitate* (JAC02138), *O. mykiss* (CAP17279.1), *D. rerio* (ABM68038.1), *X. tropicalis* (NP_001121531.1), *G. gallus* (XP_040509254.1), *M. musculus* (NP_619598.1) and *H. sapiens* (NP_055413.1).

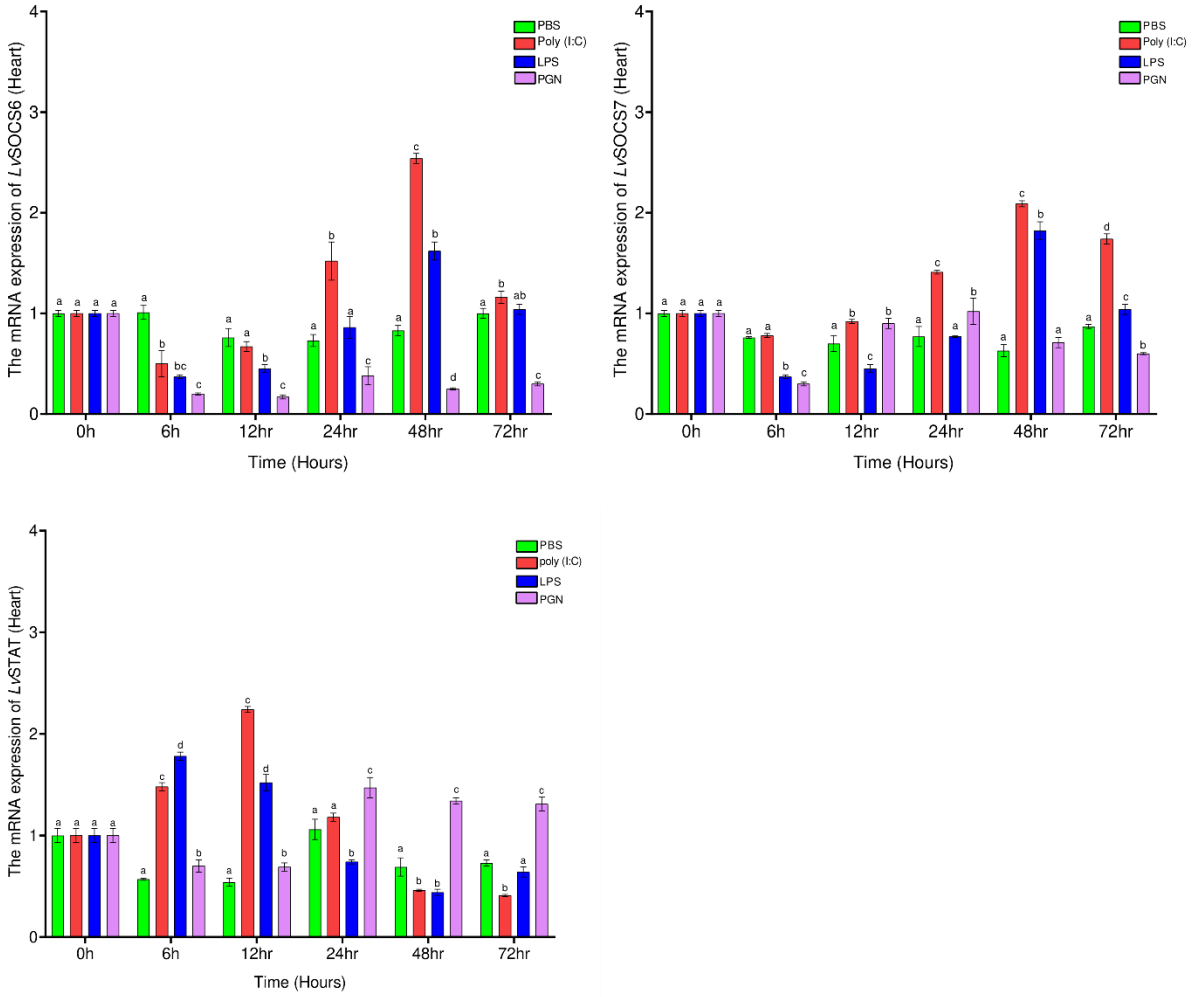

Figure S5: Temporal mRNA expression analysis of (A) LvSOCS6, (B) LvSOCS7, and (C) LvSTAT in heart tissue as determined by qPCR following stimulation of the immune response. Shrimps were challenged with LPS, poly (I:C), PGN, or phosphate-buffered saline (PBS), as a control. The expression of LvSOCS6 was normalized to EF1α internal control gene. Results are represented as the mean ± S.E (N= 3). Statistically significant values ($P < 0.05$) are denoted with alphabets.