Coleoptera Genome and Transcriptome Sequences reveal numerous Differences in Neuropeptide Signaling between Species

Jan A. Veenstra

manuscript submitted to PeerJ

Supplementary figures

Hypothenemus hampei

```
Tribolium Natalisin-R     MSTTESWKEEEGNGS---FQGFDYGLLVPISSQTPVVVDSGENFMPIWVKVLWTVIFMVM      57
Sequence-1                -----------------------------------------------------------M      1
Sequence-3                ------------------------------------------------------ILYAGM      6
Tribolium Tachykinin-R    MNFTQEFLYTTGYSIMDNSSEYDYTTNIS---YNDTEEGGNQFILPVWRQVLWSILYAGM      57

Tribolium Natalisin-R     ILIATGGNCIVIWIVTAHRRMRTVTNYFLVNLSLADLLLTTFNCIFNFSYMIQRDWPFGS      117
Sequence-1                VIVATGGNLIVIYIVLAHKRMRTVTNYFL-------------------------------      30
Sequence-2                ---ATGGNLIVIYIVLAHKRMRTVTNYFLRELKIA-------------------------      32
Sequence-3                VIVATGGNLIVIYIVLAHKRMRTVTNY---------------------------------      33
Tribolium Tachykinin-R    VIVATGGNLIVIWIVFSHKRMRTVTNYFLLNLSVADTMVSTLNVTFNFVYMLNSHWPFGE      117

Tribolium Natalisin-R     LYCIISNFIANATVAASVFTLTGISCDRYLAIVHPLQPRMSKRASLITITFIWLASMTVA      177
Tribolium Tachykinin-R    LYCKISQFIAVLSVCASVFSLMSISIDRYMAIMTPLRPRMGRTVTVLLAVTTWLLGVIIG      177

Tribolium Natalisin-R     FPCLLYSTTITNKYK-GVERTGCILIWPDGKVVGSHFDFAYQMFFLIITYVIPVTLMSFS      236
Sequence-5                --------------------------------------------------------MTYT      4
Tribolium Tachykinin-R    SPSLMFFRTYTMPYKDGEERVICYPEWPDGTTNESMMEYAYNVGFLFVTYVVPIGSMTYT      237

Tribolium Natalisin-R     YTIMGKELWGSRSIGEMTQRQIDSIRSKRKVVKMFIFVVFIFAICWLPYHGYFLYVYYDT      296
Sequence-5                YARIGIELWGSQSIGECTQRQMDNIKSKR-------------------------------      33
Tribolium Tachykinin-R    YARIGIELWGSQSIGECTQRQMENIRSKRRVVKMMMVVVIIFAVCWLPYHLYFIVISYFP      297

Tribolium Natalisin-R     DIIFSKYTQHVYLAFYWFAMSNAMVNPLIYYWMNARYVKT--------------------      336
Sequence-4                ----SAYIQETFLAIYWLAMSNSMYNPIIYCWMNARY-----------------------      33
Tribolium Tachykinin-R    EITNSTYIQETYLAIYWLAMSNSMYNPIIYCWMNARFRRGFKQFFSCLPFIHVSPGALTR      357

Tribolium Natalisin-R     ------------------------   336
Tribolium Tachykinin-R    REVLTSRRRSYSGSPDHNRIIRNG   381
```

Hycleus phaleratus

```
Tribolium_Natalisin-R     MSTTESWKEEEGNGS---FQGFDYGLLVPISSQTPVVVDSGENFMPIWVKVLWTVIFMVM      57
Sequence-1                ---------------------------------------GNQFILPIWRQALWSILYAGM      21
Tribolium_Tachykinin-R    MNFTQEFLYTTGYSIMDNSSEYDYTTNIS---YNDTEEGGNQFILPVWRQVLWSILYAGM      57

Tribolium_Natalisin-R     ILIATGGNCIVIWIVTAHRRMRTVTNYFLVNLSLADLLLTTFNCIFNFSYMIQRDWPFGS      117
Sequence-1                VIVATGGNLIVIWIVLAHKRMRTVTNYFL-------------------------------      50
Sequence-4                ------------------------------NLSIADTMVSTLNVTFNFVYMLNSHWPFGE      30
Tribolium_Tachykinin-R    VIVATGGNLIVIWIVFSHKRMRTVTNYFLLNLSVADTMVSTLNVTFNFVYMLNSHWPFGE      117

Tribolium_Natalisin-R     LYCIISNFIANATVAASVFTLTGISCDRYLAIVHPLQPRMSKRASLITITFIWLASMTVA      177
Sequence-4                LYCKITQFIAVLSVCASVFS----------------------------------------      50
Tribolium_Tachykinin-R    LYCKISQFIAVLSVCASVFSLMSISIDRYMAIMTPLRPRMGRTVTVLLAVTTWLLGVIIG      177

Tribolium_Natalisin-R     FPCLLYSTTITNKYK-GVERTGCILIWPDGKVVGSHFDFAYQMFFLIITYVIPVTLMSFS      236
Sequence-2                ----------------------------------------YNVAFLFVTYVVPIGSMTYT      20
Tribolium_Tachykinin-R    SPSLMFFRTYTMPYKDGEERVICYPEWPDGTTNESMMEYAYNVGFLFVTYVVPIGSMTYT      237

Tribolium_Natalisin-R     YTIMGKELWGSRSIGEMTQRQIDSIRSKRKVVKMFIFVVFIFAICWLPYHGYFLYVYYDT      296
Sequence-2                YARIGLELWGSQSIGECTQRQIENIKSKR-------------------------------      49
Sequence-3                ---------------------------------------------WLPYHLYFIVTSYFP      15
Tribolium_Tachykinin-R    YARIGIELWGSQSIGECTQRQMENIRSKRRVVKMMMVVVIIFAVCWLPYHLYFIVISYFP      297

Tribolium_Natalisin-R     DIIFSKYTQHVYLAFYWFAMSNAMVNPLIYYWMNARYVKT--------------------      336
Sequence-3                EITNSPYIQETYLAIYWLAMSNSMYNPIIYCWMNA-------------------------      50
Tribolium_Tachykinin-R    EITNSTYIQETYLAIYWLAMSNSMYNPIIYCWMNARFRRGFKQFFSCLPFIHVSPGALTR      357
```

Figure S1. When the *Tribolium* natalisin receptor is used as query in a tblastn search of genomic short reads from either *Hypothenemus* or *Hycleus*, the predicted protein sequences after translatation of the reads with the smallest e-values are more similar to the *Tribolium* tachykinin receptor. Yellow highlighting indicates identical amino acid residues between the short sequences and the two *Tribolium* receptors, red highlighting indicates different residues. Black highlighting indicates conceptual translation of residues that are part of an intron. Note that in all cases the short sequences are more similar to the tachykinin receptor. Hence, one can conclude that neither of these species has a natalisin receptor.

```
              1
Pogonus       MHRRW-AVVCVVYAAWASLATALPVTLLEDAKAAEAHLENKVKRSHMDESLNLNAESNSGAPVAYFSN--PSASKRA
Ignelater     ---MRSRLIWLGVVSYLTLVSSLPTSLVEDVKANEIK-DNKVKRAHSDT-------EENPRISTAYYNN--PTAIKRG
Photinus      ---MPSKLAWLGVISYLTFASTLPTSLVEDVKANSIK-DSKVKRARSDT-------EENPHLPAIYYKN--PSAIKRG
Aquatica      ---MRSNFAWLSIISYLTFANALPTSLVEDVKANSLK-DNKVKRAHSDS-------QESSHMPVVYYKN--PTAIKRG
Nicrophorus   MKHTLRALP--FLLYFAALALALPASVVEDVKSSDIK-NSKVKRAPVNAE--------S-----HGESR--PTAVKRG
Aleochara     MHPAHTGRTLAVLLTVIVVSSAIPASVLEDLISHDVA-NKKVKRAQVAVSK---ENPQPAQ--KELFY--PSAVKRS
Oryctes       MTLSRLHLPLLTVFSCILLISGLPTSLVEDVKDNEVR-NIKVKRAHDTLSR---SEKAPHDSTPYLSN--PTAIKRG
Coccinella    ---MGVQTRLLFLIFCSYGCSAIPSSMIDQIKAREMR-PNKVKRVSIAAEE---PEESLGIGNHYYGN--PTAIKRG
Harmonia      ---MGLQSRLLILIFCLYDASAIPSSMIEEIKARELR-PTKVKRVSMIAEG---PEESMKIGNHYYGD--PTAIKRG
Dendroctonus  -MAKW-AFSSLLGAYLVAVSLALPSSLVQEIKNSEMK-SNKVKRAQSD----SNEPEENIGKMSYFTNRPTSAIKRG
Hypothenemus  MNSKW-SFNCLLVASMIVLTWALPSSLIEELKNNDVNSGHKVKRGQPDSGTSGPSAEENMINTNYFDK-PTSAIKRG
Anoplophora   MGLRHVILDFLIVASSIVLVLGIPTSLVDEIKSSELR-DNKVKRAHPSDLL---TSEENGGDVPYYNK--PSAIKRG
Leptinotarsa  MGLRQVYLDLLIVATLVASAAAIPTSLVEEIKSSELR-NNKVKRAHPPNLA---SSEENEGIIPYYTK--PSAIKRG
Aethina       MD-SHFGARLFVLTSCLALVCSIPASLVEEIKSSELR-DSKVKRTPS--------NGEQNSEIQYFTK--PSAAKRG
Hycleus       MEVIWLCFKWIVWTAYLTTVLAIPGSLVDEIKANELEANNKVKRAHSI-------VEETVKDVPYYGK--PTAIKRG
Tenebrio      MELRC-SLKWATLASCMVLTLAIPASLIEEIKASELR-NNKVKRAHPPM-N---NVEERSRDVPYYSK--PTAIKRG
Tribolium     MELRW-SIRWATLASCLALSFAIPASLVEEIKTNELR-NNKVKRAHPQL-N----VGEHGREVPYYSK--PTAIKRG

             78
Pogonus       SNLNTDV---SDEAINEWAEQQQQQQQQQQQQPKIDPLKMIGYDDNPFE-DKTIAEYEKGFHYGTNKAKLDEQMENA
Ignelater     TSQ-----------TLGGWEDQS-------KESLANLAPS-LYSSDDGQFD-DKSLLDYEKAYRYGTNKEKLDEALENA
Photinus      TSQ-----------PFDNWEDLN-------KETFINLAPS-LYSN-DGKID-DKSIVDYEKAYQYGTNKEKLDEALENA
Aquatica      TNQ-----------PMENWEDLN-------KESLLNIAPS-LYSTYDGKFD-DKSLLDYDKGYQYGTNKDKLDEALENA
Nicrophorus   LNLKNGQ----LPLLTEWDQQRFRFPS--EEEPFAASP-LYTSED-ELGNDKTVAEYEKGFRYGVNKDKLDEALENA
Aleochara     EGNRLKS--SQLPLLTEWDQQRYR-----EQAERVSPS-LYSSF-------DDSPLEYAKALRYASNRDQLDEKLQNA
Oryctes       TNFND---------QIGGWDQDQ-SIFQNT----DIPSS-LYDSD---QI-DDKSIAEYEKGYRYGTNKDKLDEALENA
Coccinella    TEMKNEAADDESSDDNDWSPSEQAIYQNVDN--DFQST-VYNDE---SSQSANDMDDYDKGLQYGN-REKFDEAIENA
Harmonia      TEMKNEVPEDESSSDTDWVQSQQGIYQDVDN--DFQST-SYNDE---SSQSAKSVDDYDKGFQYGENREKFDEAIENA
Dendroctonus  TNLKEAD--------MGEWNRDQL-YGGRDFSLDNIQSG-LYDFPNLPMD-DKTIAEYEKGYQYGTNKEKLDAALQNA
Hypothenemus  TNFQDKY--------LGDDLLDFG-N-------GDDIQSG-LFN---SPLE-DKSLNEYEKGYRYGTGKEKLDEALENA
Anoplophora   TNINLKSPLPDQQSLNEWEREQ-SIFRNQAVLPNIPSS-LYNSPDDSYD-DKTIAEYEKGFRYGTNKDKIDEALENA
Leptinotarsa  ANIDLKSLP-----DLDWEREQ-ADFINRNTLPNISSS-LFQYPENNFDDDKTIAEYEKGYRYGTNKEKLDEALENA
Aethina       AN--LKNANAEQQSLSDWTQDQ-VLT---DSLSSIQSS-LYNNPNQ-YD-DKTVEEYEKGFQYGASKDKLDEALENA
Hycleus       TNQ---LPLIILSPLSDWEHEQQTFYENPENLANIQSS-LYNSD--NFD-DKTIAEYEKGYHYGTNKEKLDEALENA
Tenebrio      TN-SLKNPTPDQQSLNEWEQEQ-SLYQSPEGLANLQSG-LYNNADAPFD-DKSVAEYEKGFRYGTNKEKLDEALENA
Tribolium     AN-NLKNPSPEQQSLSDWEQEQ-SLYQNPDSLADIQSS-LYN-AENPFD-DKTIAEYEKGFHYGTNKEKLDEALENA

            155
Pogonus       ILKSEMYGDPASI-NQYRYYGGSDDRRRKRSN-----------LKRSASFANRYKREVELSPEDILTLLALWDEDRLR
Ignelater     VLKSELYGDPGAL-NQYRYFDGSSDERRR-KR----------RAIKNLRLNSRYKREIELTPEEVLTLLTLWENERRR
Photinus      VLKSELYGSPGSA-NQYRYFEGGNDEKRK-KR----------AAF---KRLNSRYKRDIELTPEEFLTLLSLWENERRH
Aquatica      VLKTELYGNPASF-NQYRYFDADER--RK-KR-----------GT---YKLKDRFKRDIELTPEEFLTLLSVWENERRL
Nicrophorus   ILKSELYGEPAAV-NQYRYYGMEGD--KKRKR----------RNAQKTRMDNRYKREVDLSPEDILAILTLWENERQH
Aleochara     ILKSEMYDLPATLPNQYQYYGNDDRRRKRRSTNGKLAKKAARVQKARSITNRLKRSADMSPEQILAILTLWENERMN
Oryctes       VLKSELYGDPVSI-NQYRYYGGVADEKRRRRR----------SNAQKMRFDTRIKRDVDLSPEDILTLLSLMENERQR
Coccinella    VLKSEFYGNLEGD---------------KKKRKK----------RNSRQYSYGNS-AASDDLSPEEVLDLLTLYESERQK
Harmonia      VLKSEFYGNLDND---------------KKKRKK----------RSSRLNGYGNS-IASDDLSPEEVLELLNLYEGERQK
Dendroctonus  ILKSELYGDNY---GPSIWPYEDRRRR---KR----------ETVSNKRNTLRYKRNVDLTPDEVLRLLQFYEKNQRE
Hypothenemus  ILKSDLLLDPY---RRYN---------------------------NRSKRTVDLTPEGMAALLSLYEKNHR-
Anoplophora   VLKSELYGEPGAI-NQYRYYGADD--KKR-KR----------REVKKLRFDGRIKREVDLTPEEILALLSLYESNRQR
Leptinotarsa  VLKSEIY-EPTSI-NQYRYFGAEAA-KKR-KR----------RDAKKLRLESRMKREVDLSPEEILTLLSLYENNHPR
Aethina       VLKSEYYGEPGAV-NQYRFYGGDD--RKR-KR----------RSAKKLRLSQRLKRDVELTPEEILTLLNLYEKSRQN
Hycleus       VLKSELYGEPGAI-NQYRYYGNDDK-KRRRRR----------RDVQKSSRTNRLKRDTDLTPEEILSILTLYENEHQN
Tenebrio      VLKSELYGDPAPL-NQYRYYENDDR-RRR-RR----------RDARKIRPDNRIKREVDLTPEEILTILTLYENERQS
Tribolium     VLKSELYGDPAPL-NQYRYYGNDDQ-RRR-KR----------RDARKIRLDSRMKREVDLTPDEIFTILTLYENER--

            232
Pogonus       Q---PAGYRTNWPRYNPDIDLDNDREE---NEIDEPEESWLESPV-YPGIRNP-VHAS--PF---ARE--YAHNQVQ
Ignelater     A---QDDYRPTWSRYEPNLDLDDSSEN---D-VEQEDENWLDTPVVYPHATNR-INQLSGEFVRGNPNYIYEDK---
Photinus      T---SDDYRPTWSRYEPNVDLDDRNEN---E-LDEDDENWLDTPVVYPHATNR-----------ASPNYMYEDK---
Aquatica      S---QDDYRPTWSRYEPSVDLDDRNEN---D-VDQDEENWLDTPVVYPHATNR-----------GNPNYIYEDK---
Nicrophorus   GPP--ANFHPSWQRFD-NFDMDSISGDKNEPEIEQNDEDWLDSPV-YPHISEH-LSPM-------NPNYYY-DS---
Aleochara     K------DATPWLNYDEDGYGD---------MSSTDQDEDWLEAPV-HPHATDE-H-------MMPQSPNYYYDVSPQA
Oryctes       P-----SNYRNWQRYDSNLDLDKDNQD---SYEEEDEGTWLDTPV-YPHVGPH-TNSM-------PSSN-YYYE---
Coccinella    P----------------EDTL----------SNLEQDNSIWLDVPV-RPGAILPSTSNF-------GPAYHLDRSPSN
Harmonia      P----------------EMDI----------GNFDQDHAVWLDVPV-RPGPVVPNTSNL-------GPSYSLDRSPLD
Dendroctonus  LK---------------NGEFY---------PVEDNNDETWFNEPMRYG-SV-D--RP------------ANNRFHQ---
Hypothenemus  -S---------------D-PYE---------IADEDEDGAWLNEPVVRPHSSIE--NS------------VYNTRNL---
Anoplophora   QQE-SNNYRQPWNRYEPNFDVDINQDD---S-QDQDEENWLDNPV-FPHATGY-DKEL-------GPKYMIDQS---
Leptinotarsa  QQPSTENYRYPWNRFEPNLDLDINQDD---S-QDQDGENWLDNPV-YPHATAF-EKDL-------GPKYMYEPS---
Aethina       D------NYRPWQNRLEN-SFDNFEDG---E-LNDNDESWLETPV-YPHVSRD-NTDI-------GPQYMIDE----
Hycleus       ---------RPWRLDDF--------DK---S-NEQDDETWLDGPV-YPQTNG---NDI-------GPGYMFD-----
Tenebrio      N------GYRPWSGEP-D-----NDQN---N-NIEEEEENWLDAPV-YPHAAGH-TNDI-------GPSYLLDE----
Tribolium     N------GYRPWGLEP-E-----PS-G---D-NLEEEEENWLDAPV-YPHATGH--NDL-------APSYLMDE----
```

```
                309
Pogonus         FEQPPIFAQKRQWAGF-SD-KKRKRFMVARKR-SDPTRELRYLNGPAQSHEDLYSLTQLLGPPPARD---GNFPMYH
Ignelater       --RS----QLGQWGGF-SD-NKKKRFMVARKR-NDPTRELRYFNGPAQ-RNDFYTLSQLLA--SQRE---PNVPLYH
Photinus        --R-------GEWGRF-PE-NKKKRFMVARKR-NDPTRELRYINGPPQ-RSDFYTLSQLLA--AQRE---PNAPLYR
Aquatica        --R-------GQWGRF-SE-NKKKRFMVARKR-NDPTRELRYINGPPQ-RNDFYTLSHLLA--NQRE---PNAPLYH
Nicrophorus     --KPQVYEKRRMWGEY-PD--KKKRFMVAKKS-NDPTREIRFLNGPT--RNNYYTLSELLGA-AQQE---QNVPLYQ
Aleochara       PPVPQFYEKRPRWGGF-NE-NHRKRFMVAKKR-GDPTRELRYLNGPS--HSDYYTLSELLA--NQRD---PDVPLFQ
Oryctes         --KPQSYEKHNQWNEV-AE-SKKKRFMVAKKR-NDPTRELRYLNGPN--VNDYYTLSQLLG--NQRE---PNIPVYH
Coccinella      FEK-------SRWS----DTRNNKRFMVPKKR-NDPTREIRYINGPT--KHDYYFLSQLLN--NRRE---TNVPVFH
Harmonia        YER-------SKWL----ENK-NKRFMVAKKK-NDPTREIRYLTGPA--KHDYYFLSQLLN--NRRE---ANVPVFH
Dendroctonus    --INSQPDYKPRWGAFDPATFKKKRLFY-------------------------------------------------
Hypothenemus    --LDDIYHLKPRWGSN-IDFNKYKRFPMAKKRTTDPTRIIRYLNGPS--QNDFNTLSSILN--SQKEI-EIGVPVYR
Anoplophora     --NSQMFDKKGRWGGF-GD-NRKKRFMVSKKRTNDPTREIRYLNGPN--KNDFYTLSQLLS--NQRE---PNVPVYH
Leptinotarsa    --GPQVFENKEKWRGF-GGDSRNKRFMVAKKRNVDPTHELRYINGPN--KNDFYTLSQLLS--NQRE---PNLPVYH
Aethina         ---PQLYDKRGKWGNF-AD-NKKKRFMVAKKR-NDPTRELRYINGPN--QNDYYTLSRLLS--NQQE---PDLPLYR
Hycleus         --------DKRRWGNL-VDNERKKRFMVAKRR-----------DDPA--RNNFYTLTQLLS--NQRE---PNVPVYH
Tenebrio        ----KPFEKRGRWGGF-AD-NRKKRFMVAKKR-NDPTRELRYLNGPN--KNDYYTLSQLLS--NQRE---PNVPLYH
Tribolium       --------KRGRWGGF-AD-SRKKRFMVAKKR-NDPTRELRYLNGPN--KNDYYTLSQLLS--NQRE---PNVPLYH

                386
Pogonus         RMVL
Ignelater       RFVL
Photinus        RMVL
Aquatica        RMVL
Nicrophorus     RYVL
Aleochara       RYVL
Oryctes         RRVL
Coccinella      RYVL
Harmonia        RYVL
Dendroctonus    ----
Hypothenemus    RPIL
Anoplophora     RLVL
Leptinotarsa    RLIL
Aethina         RLVL
Hycleus         RLVL
Tenebrio        RLVL
Tribolium       RLVL
```

Figure S2. Sequence comparison of Coleoptera baratin precursors.

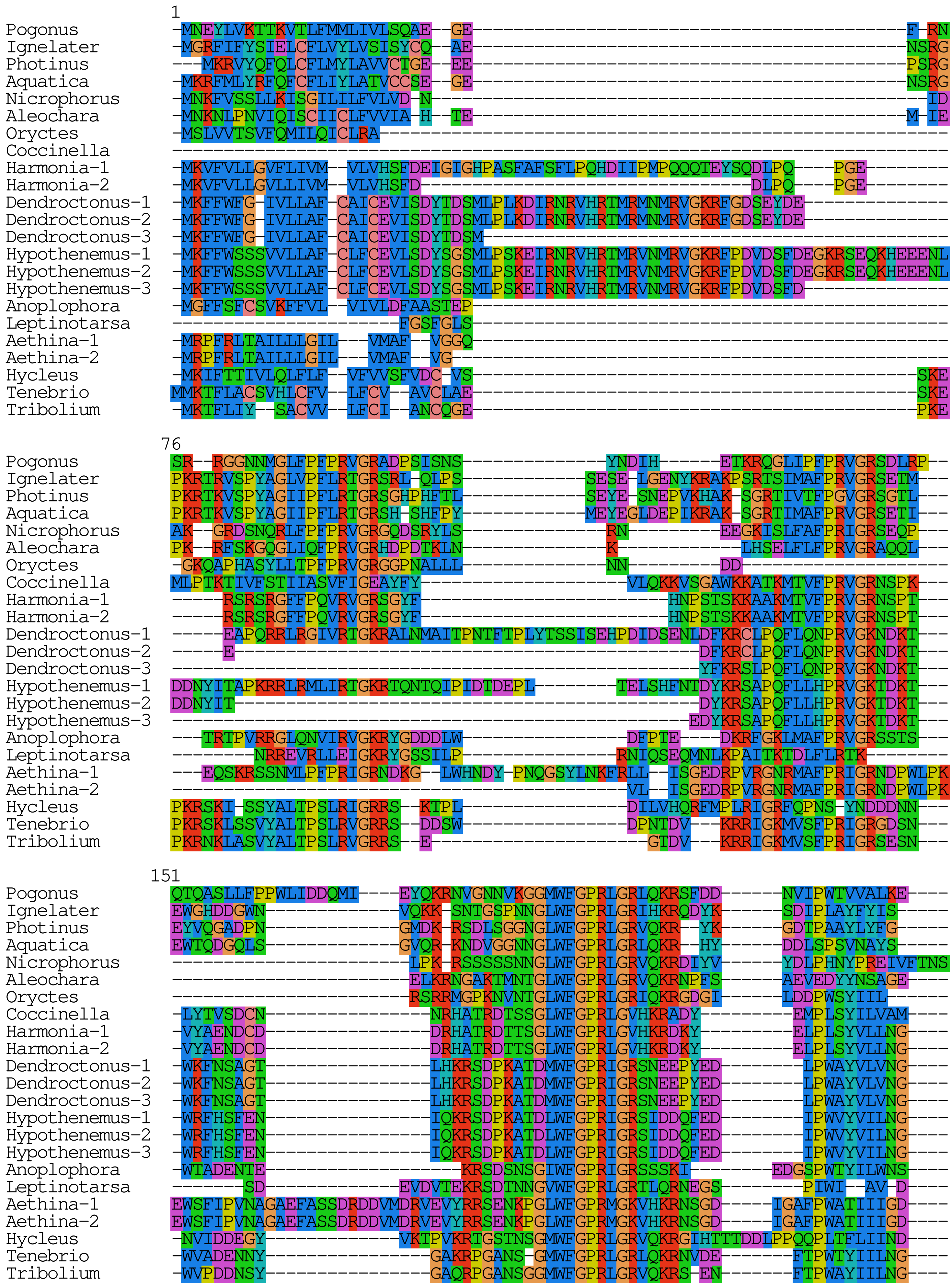
```
               1
Pogonus        ---------MHSCT-LILITFGIIAVLMTCSSAS----AIPQPPCSPSVLMT-ADAKYRQICS
Ignelater      ---------MHQCT-ILIASVGILAAFLTSSSVT-AS-VDTTNLCPPDASP-----KMRQLCL
Photinus       ---------MHQYK-IIIASICIFAGLIS STVN-LS-VETTNLCSSDASP-----KLRQLCL
Aquatica       ---------MHRYT-ILVASVGVLVALLTS SVA-IN-VETTNLCPSDASP-----KMRQLCL
Nicrophorus    ---------MNQCT-VYAVAF IFVAMMASSNLS-MASNLPLIYCQPSALD---GLQKRQLCF
Aleochara      ---------MHRESSYFALGVGLLVVILAARTTTTSAAHIPVIVCPPNPNSADVPSQLRQLCF
Oryctes        ---------MNYST-DIIATFGIISIFLISSTSS---TTIPMINCEPNSLS-EAPTKIRQLCY
Coccinella     ---------MHLYI-TSALLWGVLSILMH--------SSSASVISCPPNNLP-EAGVYLRQLCS
Harmonia       ---------MHLNF-VAIFFFGVLATSML--------STSASVVSCPPNSLP-EAGIYLRHLCS
Dendroctonus   ---------MQQST-VYF FFGMLVFVLLC-----SMTSATVISCPPNSIQQEINPKLSQLCF
Hypothenemus   ---------MHQSV-VCF FFGMVVFAFFY-----SVATASVISCPPNSVQQEINPKISELCY
Anoplophora    ---------MQTYT-VFSLIFGVVTLILTS-----SISKASVISCPPSSFQEQSNPALRQLCY
Leptinotarsa-1 ---------MHSST-IVSCVVGTIIVILFS-----SVARASIISCPPNPIQEQVHPALRQVCD
Leptinotarsa-2 MFTHTNSQMNSFM-MVSFIFWM--AVLS------STSQAYVINCSPVSVQDQMNPGVRQLCA
Leptinotarsa-3 ---------MHFSA-VISFVVSLVSAVLS------SEVNAYIISCPLSSLQDTSNSGLRQLCS
Leptinotarsa-4 ---------MDFLL-KIYFLVSL--MFGI------NVSNAYIINCSPSTLQIQANSRERQLCA
Aethina        ---------MQQHT-FAIFVLGVVSVFLSC-----STSTASVISCPPNNL--EVAPNLRQLCY
Hycleus        ---------MHQYT-FFII FGFVAVFVSN-----SLITASIISCPPSNLQ-EANPNLRQLCY
Tenebrio       ---------MHHYT-LVAV-AFAAVAALLT-----SSATATAISCPPNPL--EASPYVRHLCY
Tribolium      ---------MQQYA-FVAIVFGAVAVFLAN-----ASTTAYYVSCPPNDAL-EASTSLRHLCS

              63
Pogonus        LLSQ--FARDLEFSSDDRV------PDMSELI-------DNGIKRKDVDHVFLRFGRPRM--
Ignelater      LASLEQFLAENPP-RPNAD------VLDTQLY-------DRGTKRQDVDHVFLRFGRRLGL-
Photinus       LASLEQFLSENPS-RSNAD------VQDTQLY-------DRGTKRQDVDHVFLRFGRRLGL-
Aquatica       LASLEQFLAENPP-HSNAE------PMDTQLY-------DRGTKRQDVDHVFLRFGRRLGL-
Nicrophorus    AL-LER  MDAPQ-EVSND------VMDNQLY-------ERGIKRQDVDHVFLRFGRRMGL-
Aleochara      VL-LDKNDIQIPLEDGDLAAAGIDPAQAAQLYERHLVEGSRGTKRQDVDHVFLRFGRRSGM-
Oryctes        YV-MEQLRSEQ P QDNGE------MIDSKLY-------ERGIKRQDVDHVFLRFGRRFGF-
Coccinella     AI--KQAINDNSPPEEPYY--------F-----------GGNSKRQDVDHVFLRFGRRLGFE
Harmonia       VI--KQAINDNAPPEEQFY--------F-----------GGNSKRQDVDHVFLRFGRRHGFE
Dendroctonus   AI--EQALQDSSPQNDAYT--------ARLVDERNT---NLNAKRQDVDHVFLRFGRRFGM-
Hypothenemus   AI--QQTLQDSSPQNDEYI--------GQLLDERTASNLRTNAKRQDVDHVFLRFGRRYGL-
Anoplophora    AI--EQAVDEVVPQQTQFP--------GRLEE    RNTNLNAKRQDVDHVFLRFGRRFGL-
Leptinotarsa-1 AI--EQAIVDI SQQQAFV--------GRLEE----RNTNLNTKRQDVDHVFLRFGRRFGV-
Leptinotarsa-2 VL--DSTID LTS PPAYS---------GLLG-----ESISLPNDKKLDHSFLRIGKRFHL-
Leptinotarsa-3 IL--EQAVDDLPQ-QAVYS---------GIVG-----DSNTLSSGKRVDHSFLRIGKRFNF-
Leptinotarsa-4 LF--DPAID-LVA-QTAYS---------GILG-----ESVTLPNDKRVDHNFLRIGKRFNL-
Aethina        AI--EQAISEVQNNQAEYH--------FDERN----APGQVEKKRQDVDHVFLRFGRRLGY-
Hycleus        AI--EQAISENAISQDQ F--------GRMDE----RNVNPNAKRQDVDHVFLRFGRRFGL-
Tenebrio       AI--EQAISENAITDDQ Y--------RRVEE----RNVNGNAKRQDVDHVFLRFGRRLGL-
Tribolium      FI--EQAVNDNVVIQDEPF--------RRIVG----RNVNPNTKRQDVDHVFLRFGRPFGL-
```

Figure S3 Sequence comparison of Coleoptera myosuppressin precursors.

```
Aquatica      HLNGRTLDELGGGHLIG-RTLDELGGGHLNG-RTLDELGGGHLNG-RTVDELGGGHLNGRNAY
Nicrophorus   NLNGRALDGIGGGNLNG-RALDGIGGGNLNG-RALDGIGGGNLNG-RALDGIGGGNLNGRSYL
Harmonia      NLRKRALDSIGGGALLGKRSLDSIGGGALLGKRALDSIGGGALLGKRALDSIGGGALLGKRAL
Coccinella    MLRKRALDSIGGGALLGKRALDSIGGGALLGKRALDRIGGGALLGKRALDSIGGGALLGKRSQ
Dendroctonus  LASRRRLDSIGGGGILSARGLDSIGGGGLL--RSIDDIGGGGILSARSMDGIGGGGILEARSM
Anoplophora   TLMGRGLDGIGGGNLMG-RSLDGIGGGSLIG-RGLDGIGGGTLMG-RGLDGIGGGNLMG-RSI
Leptinotarsa  SLIGRSLDNIGGGALIG-RSLDNISEGSRIG-RSLDPIGGDSLIG-RNLESIGGAVKRTIDNI
Aethina       LYRAPTMDDIGGD-VL-KRSMDDIGGD-VL-KRSMDDIGGD-IL-KRSMDDIGGDVLRSLYRA
```

Figure S4. Sequence alignment of parts of the orcokinin B precursors from the species indicated. Note that the putative convertase cleavage sites consists of a single Arg residue in most species, but that in the two Coccinellids, Harmonia and Coccinella, all have been replaced by a Lys-Arg pair, while in Aethina several single Arg convertase cleavage sites have similarly been mutated into Lys-Arg doublets.



```
                  226
Pogonus           ----------PGKQYDYVPRSGREDEEEDNDDTVVET----VLSRRLDSKTRNNISQQ
Ignelater         ---------------DYTPRLGRESEETMDETDTIS-EDQVNNDKTMLESA-------
Photinus          ---------------EYTPRLGRESAEEIEDD----GEDESNLNKSM-----------
Aquatica          ---------------EYTPRLGRESEEVLQNIEGASDDDDVDLNKSI-----------
Nicrophorus       KYNMDFPDYPETRNVHYTPRLGRESSEQQEEDAEE-----------------------
Aleochara         ----------------------------------------------------------
Oryctes           ---RDVPAYQ--HIPKYTPRLGRESQENMDDNAEDMN----QASSS    DRA-----
Coccinella        ---EHGLNTSLTLIRTSIPMVKMKNWITDLS-------------------------
Harmonia-1        ---AQGTRMEYSPFPD-------RDFESDDGSEKL-D------FESYN----------
Harmonia-2        ---AQGTRMEYSPFPD-------MDFESDDGSEKL-D------FESYN----------
Dendroctonus-1    ---NRPING--------A-VFK--SNEVEDQPRDDLPE--------------------
Dendroctonus-2    ---NRPING--------A-VFK--SNEVEDQPRDDLPE--------------------
Dendroctonus-3    ---NRPING--------A-VFK--SIEVEDQPRDDLPE--------------------
Hypothenemus-1    ---NRLQSN--------IPLLK--SNDDQEQSRNESGN--------------------
Hypothenemus-2    ---NRLQSN--------IPLLK--SNDDQEQSRNESGN--------------------
Hypothenemus-3    ---NRLQSN--------IPLLK--SNDDQEQSRNESGN--------------------
Anoplophora       ---D----KPLNRHTHDINSR---GDGEEDSSSENNGEESKEQLFNY-----------
Leptinotarsa      ---E----YPMEQNRHSNSRN---TLSEEEPLEEVAYE--------------------
Aethina-1         ---AHVPPKFF-----Y-------NNPAAEPSTNDLDKP-KDQN--------------
Aethina-2         ---AHVPPKFF-----Y-------NNPAAEPSTNDLDKP-KDQN--------------
Hycleus           ---DGTNDGLLNRQVHYTPRLGRESEELLIDDSDNT-MA-------------------
Tenebrio          ---E----GPVSRQVHYTPRLGRESDEVYDEL DAD-VD-VLA---------------
Tribolium         ---E----APIIREVHYSPRLGRESEEAYEEILDSN-LD-VL----------------
```

Figure S5. Alignment of periviscerokinin precursors.

ARCHOSTEMATA

Micromalthus debilis (GDOQ01020265.1)
MALLWALVLFPAAILASGTEQEPGIYPQYYSRYPDREEPPFAFPEEFQSDQRKTVQKSYEEIAEDLSKLSNNLQ
QKLRFASALLKQENPYETDPADRIEISNLQNSLCILNAEIDNYFRQKAGVFSKGNTQAKRSVWDSKKSTFDSAG
RTSPIELANKSPNIYKSNSNLANAYAQPLFTMKFDATPTDGRTPVQRKRREITKAQLENLLQDEIEKQEEEEWE
RIEGEDDFKKRSQNPPTVVLKSGEVDIPKRVPFNPWGGKRLEQFPVNRDLLGVWESKRDRMPFDPWGGKRARVP
FDSWGGKRDRTAFDSWGGKRDRLPFDSWGGKRSSRVKLSPWAEGKRPKQRKSQFYSWGGKRSKYDATLV*

Priacma serrata (GACO01006445.1)
-----GGDDTDQKRTKFFSWAGKRTNDGTEPSFEFLPSFKRSHSSPNIVVKPREMALPKWTPFHSWGGKRAVQV
ESERLPISRVPFNAWGGKRSSDHEDNEFLPLHQEMTSLPHNPEGEFFKSVQTIETEVEVSQLGWKEKSVLTLRN
IKMLPL*

ADEPHAGA

Pogonus chalceus (JU438197.1)
MYYKNSVIQTSKILLQIILYILIQFANTNCMTIPNDNSNMLQTKSIRNRLNLLEDDNIFGTETPTDTKTYNSGN
DMENNNNGDIFTKETDNENIFTEDSDIDDKKLNVNKIVSLLLDPNEVTPKMIEKLSQFKTFYLSNSNDKTLINL
CTLSRNYINDVCLLIQSSNRSNGLYNPYIYQRCVKLLPLLDFYHCVNEKQNYDETAKENEFKRVPFHAWGGKRF
RQVATEKVQSSKVPFNAWGGKRQMNYDDGDEYCKRSKSKFHSWGGKRTVRETANVL*

Gyrinus marinus (GAUY02012896.1)
-----QMLMDASKQVDDEDAKHVKFLGWIGKRFSNANGNNIPKRIPFHAWGGKRSNQVGTEKIQASRVPFNAWG
GKRTYNMNSDESTPDNMYEINTPSSVLPYLVYKRSRTKFHSWGGKRNY*

Figure S6. Predicted partial or complete Coleoptera leucokinin precursors as obtained from conceptual translation of TSA transcripts found at NCBI. The nucleotide identifiers are indicated in parentheses. Lys-Arg and Gly-Lys-Arg processing sites are indicated in purple and the amino acid sequences of the mature peptides are in green.

```
                  1
Ignelater         NLEDLASLNLEQIEMDNVDVSIFSRLNKLESVYFKKYHYCAFAPTVPRCRPLTDGVSSAHHLLEKPVLRYATWI
Photinus          KMENLQSLNLDGIEIENIDLSIFSTTPNLKSVYFKKYFYCTFAPRVRRCRPLSDGISSVDQLLAKPILRYAIWI
Aquatica          NLKNLESLNLEGIEIHNIDVSIFSSLKRLKSIYFKKYFYCTFAPSVKRCRPLSDGISSVDELLAKPILRYALWI
Dendroctonus      NLQNLLSLHLEEIDITNIHTDIFASLTNLQWIYFKTYKYCTYVPTVPRCRPLSDGISSINQLLFKPIFRYSNWI
Hypothenemus      NLPNLLSLHLEDIEILNIDTKMFGGLDGLKWIYFKTYTYCTFAPEVPRCRPLSDGISSLNQLLYKPIFRYSNWL
Anoplophora       NLINLNLLNLEDIEIRNIHTLMFINLFSLESVYFKKYSYCLFVTSVERCRPLSDGVSSNAQLLYKPIFRYTNWI
Leptinotarsa      NLTQLQHLNLGDIDILKIDASMFDSLKELKRIELKNL-ICRYN-KVLSCR-ASDGISSANNILNKGIFRFWNWV
Aethina           NTINVRSLNMEGIEITNIDLGMFEHLSDLQSVYFLKYSYCSYSPMVPRCRPLSDGISSTKQLLYKPILRYSNWI
Drosophila-LGR3   EPLNISFLNLTGIRYDHIDFEAINSMRNLTYIIYDRFFYCSMTPRVRMCKPSTDGVSSFQDLLSKPVLRYSAWV
Drosophila-LGR4   CLKNLSSLLLTGNRFQTLDSRVLKNLTSLDYIYFSWFHLCSAAMNVRVCDPHGDGISSKLHLLDNQILRGSVWV

                 75
Ignelater         TCLIACAGNISVIWGRLTV-RDENRVLSFVVRNLAASDLLMGIYLLIIGFHDVKFRNIYNKVSHQWMSSWLCTT
Photinus          TCFVTCIGNAMVIYERMTS-RDENKVLSLVIKNLAASDFIMGIYMVIIAGYDLKFRNIYNKVAHEWISSWPCTF
Aquatica          TSLFTCVGNAMVVCGRVMY-RDENKVLNLVIRNLAVSDFLMGVYMIIIAFHDFKFRNIYNKI-HIWMSSWFCTA
Dendroctonus      MCIITIGGNSLVLFGRFLF-RDENKNLSVVIKNLAVSDGLMGIYLLIIAYHDLKYRDNYNEIARKWMSSWNCVI
Hypothenemus      MCVITIGGNSLVLFGRFLF-RDENKNLSIVIKNLAVSDGLMGIYLLSIAYHDLKFRDNYNEMARKWISSWTCVF
Anoplophora       ICFATCAGNIMVLFGRYLF-RDENKVLSLIIKNLAVSDLLMGIYLLLIGIHDMKFRNIYNLEAQNWISSWGCTI
Leptinotarsa      ISSAICISNFLVIFSRLFY-RDENKILNLVIRNLAASDFLMGVYLIIIGIYDVKFRGVYGEMSEQWLSSWTCTF
Aethina           ICFITISANTLVLFGRYMF-RDENKALSLVIRNLAISDLGMGIYLLIIAVQDWRFRNAYNQYAHAWISSWGCTV
Drosophila-LGR3   MATLTIAGNVLVLWGRFIY-RDENVAVTMVIRNLALADMLMGFYLVTIGVQDYRYRNEYYKVVLDWITSWQCTL
Drosophila-LGR4   MASIAVVGNLLVLLGRYFYKSRSNVEHSLYLRHLAASDFLMGIYLTLIACADISFRGEYIKYEETWRHSGVCAF

                149
Ignelater         AGILAMTSSEVSVLLLVFMSVERCLLITAPFGHLASLTLKPAFSILFSIWTVGIGLAVIPVIEFYS-STR-FYG
Photinus          AGVLAMVSSEVSVLLLVFMSVERFLIIAVPFGRFSSIDVKKCALILGCIWILGVGLAIIPVLQFYS-STK-FYG
Aquatica          AGVLAMVSSEVSVFLLVFMSVERLLIIAVPFGPFSSMDLTKTVTVLXSIWILGIGLAVFPILQFYS-STR-FYG
Dendroctonus      TGIIGMTSLEVSVMILVFLSVDRYFVITMPYKKYGSLTLKETWRVILCIWCFGICISVIPALGLLS-STK-FYG
Hypothenemus      TGVVGMISLEVSVMFLVFLSVDRYFVITMPYKKYGVLSLKETWRIVFFIWCFGISISVIPVLGFSS-STK-FYG
Anoplophora       TGIIAMLSCEVSVLLLVFMSVDRFLLIAIPYGKYSALTMREAVIALLFIWCLGIILAVFPALAYLS-STR-FYG
Leptinotarsa      TGITAMISSEVTVFILLFISIDRYLVIVTSYGKYKSLNEKDTVIVLTIIWCLGIAISVIPVIRYAD-DTK-FYG
Aethina           TGMIGMLSSEVSMLLLLFMSIDRYLLIAVPFGRYHALTLRKTTLILIGIWLVGVFITVLPVIEYHN-STR-FYG
Drosophila-LGR3   IGTLAVSSSEVSMLILAFMSLERFLLIADPFRGHRSIGNRVMWLALICIWITGVGLAVAPVLLWRT-STLPYYG
Drosophila-LGR4   AGFLSTFSCQSSTLLLTLVTWDRLMSVTRPLKPRDTE-KVRIVLRLLLLWGISFGLAAAPLLPNPYFGSH-FYG

                223
Ignelater         -VNSLCFPLHISDPFFLGWKYSAIIMLGVNSASLIIIAVVYCVMFISIWKTRHATSLP--LRDFEFAIRFFFIV
Photinus          -LNALCLPLHTTQAYHVGWEYSAFIVLGINFVSLITITLVYCIMFISILRTRSATTLP--RKDYEFAIRFFFIV
Aquatica          -LN-LCVPLYITEPFFVGWKYSAFIILGINFISLIVITVVYSVMFISIAKTRNATTLP--IKDYEFAVRFFFIV
Dendroctonus      -VNGLCLPLYIDEPYLIGWEYSAIIFFGINASSLLIIIYTYGGMFINIKNTRNATSMP--MNDCQFAIRFFFIV
Hypothenemus      -VNGLCLPLYINDPYFIGWQYSALIFFGVNASSLIIISYTYARMFINIKNTRKATSVT--LNDGQFALRFFFIV
Anoplophora       -VNGLCFPLHIDDPYLEGWEYSAVIFFGINATGLIIISVVYIGMFISIWRTRNATNLP--ANDYQFAIRFFFIV
Leptinotarsa      -INGLCFPLHIAYPYYSGWQYTAFIIFGINATSLIIIAFVYIGMFISIRKTREATPLP--KKVYEFVLRFFFIV
Aethina           -VNGLCFPLHIDDPYFIGWEFSVTIFVGLNAFCMIVTTVVYIGMFVSIWRTRNATTLP--SKDYEFAVRFFFIV
Drosophila-LGR3   SYSGTCFPLHIHEAFPMGWLYSAFVFLGVNLLLLVMIAMLYTALLISIWRTRSATPLT--LLDCEFAVRFFFIV
Drosophila-LGR4   -NNGVCLSLHIHDPYAKGWEYSALLFILVNTLSLIFILFSYIRMLQAIRDSGGGMRSTHSGRENVVATRFAIIV

                297
Ignelater         LTDASCWFPIILVKVFALTGMEISADLYAWLVVFVLPVNSAVNPLLYTFTTPKYRSKINKVAVPLHCIRDYGNM
Photinus          FTNGCCWFPSVLVNVLLLLGKEISGDLHAWLVVFILPINSAINPILYTFTTPKYRFNSKKVLVFFNMISQSQRS
Aquatica          LTNGCCWFPIILVKCLVFFETEIPNDLYAWLVVFVLPINSAINPILYTFTTSKYRSKLQKVNIMLHWILKNERS
Dendroctonus      LANIACWVPIITAKMLVYLKVDISGDFYAWLVVFVLPINSSLNPILYTFTTRKYRRRILKVPSCLKKFTTVNNL
Hypothenemus      FTNVACWLPIILAKMLVYFTVEVSADFYAWLVVFILPINSSLNPILYTFTTRKYREKIVNMPQCLRKCAYQSRP
Anoplophora       LTDVTCWLPIVTIKAVAFLGVAVSDDLYGWLVVFILPINSALNPILYTFTTPKYRTQILKKVPSLIKEAAVNLS
Leptinotarsa      FIYGLCWLPIIVFKTAAYAGADISDELYGWLVVFILPINSASNPILYTFTTRKYRAKMKISCIFEGVMDRTHHE
Aethina           LTELFCWLPIITIKVIVLFQFSVSDDTYGWLVVFILPINSGLNPILYTFTTPKYRREIYRVPMSVRESTSNKTR
Drosophila-LGR3   LTDFLCWVPIIVMKIWVFFNYNISDDIYAWLVVFVLPLNSAVNPLLYTFTTPKYRNQIFLRGWKKITSRKRAEA
Drosophila-LGR4   TTDCACWLPIIVVKLAALSGCEISPDLYAWLAVLVLPVNSALNPVLYTLTTAAFKQQLRRYCHTLPSCSLVNNE

                371
Ignelater         VLSDNIRRQSPQSHL--------------------------
Photinus          SLEKTTQGQTVPSSGGAFLTSSSI-----------------
Aquatica          NLKKNSRHPPVPSSAGAILNSSSL-----------------
Dendroctonus      TETHLPGAIFKITQKDIKTVNN-------------
Hypothenemus      SETQLSREIFTISQRDIEREKYF------------
Anoplophora       KNNGSKICSGEIDHLKLTCAKENNLNKTNVKIVLNKF
Leptinotarsa      DPSLTDPSDVESDCGVSKSKSTTEKRF------------
Aethina           TYDKGSPGTYHSVFDLKKFLQYQTPFVQLI----
Drosophila-LGR3   GNGNVATTTTGTATGSSQHPDDFTIFAKAAMRCH----
Drosophila-LGR4   TRSQTQTAYESGLSVSLAHLGGGVGGGSGRKRMSHRQMSYL
```

Figure S7. Alignment of N-terminal parts of Coleoptera LGR3's; for comparison the LGR3 and LGR4 from *Drosophila* have been added.

```
                   1
Pogonus-EH1        -------MQTKDSYHKIVLIFFV-LFVMIIMTSANPAIDYFPKPFIYTKNYGICIRNCAQCKKMFGPYFEGQL
Ignelater-EH1      ---------MKI----FLQICILCF-STVLLTEANPI--------------GVCIRNCAQCKKMFGPYFEGQL
Photinus-EH1       -----MACSMKT----IFRIYFFCI-AALPFIEANPI--------------GICIRNCVQCKKMFGPYFEGQV
Aquatica-EH1       -----MSRTLKT----VLRLYLLCF-ATFLLTDANPI--------------GICIRNCVQCKKMFGPYFEGQL
Nicrophorus-EH1    -----MVSSRKFIVLTGAALLLLLVVSQSPVVEANPI--------------GICIRNCAQCKKMFGPYFEGQL
Aleochara-EH1      -----MGFLLRRGFEST-FCLVVLFAALVLQAQANPI----------GILHGVCIRNCAQCKKMFGPYFEGQL
Oryctes-EH-1       -------MAKTVYLG-----IFLSTLLLATFVNSSP------------LFTGICIKNCAQCKEMYGKYFYGQL
Coccinella-EH      ------------MLRKV--ILLVFICCMIVHLEAYNP------VTKLSRPIGICIRNCAQCKKMYGPYFDGEL
Harmonia-EH1       ------------MLRIT--VLFVFVCCMFVHLEAYKT------GSRNTKPIGICIRNCAICKKMYGPYFDGEL
Dendroctonus-EH1   -------MAEYQRIT---LRILMVVVLAASCIDANLI--------------GVCIRNCAQCKKMFGRYFEGQL
Hypothenemus-EH1   ------------MLP---TFILTVLMFSAVCIEANPI--------------GVCIRNCAQCKKMFGPYFEGQL
Anoplophora-EH1    -----MVCQRNKMLG---VVAFLLLASMVLLAEANPI--------------GVCIRNCAQCKKMFGPYFEGQL
Leptinotarsa-EH1   -----MEGGVKMYSG---VAVVLIFTCFVFCAEANPI-------------VGICITNCAQCKKMFGPYFEGQL
Aethina-EH1        -----MVRVAKNLLG---ASFLFLIASTILFADANPI--------------GVCIRNCAQCKKMFGPYFEGQL
Aethina-EH1        -----MVRVAKNLLG---ASFLFLIASTILFADANPI--------------GVCIRNCAQCKKMFGPYFEGQL
Hycleus-EH1        -----MVNTTRT----SLIICLLLINNYLILAGANPI--------------GVCIRNCAQCKKMFGPYFEGQL
Tenebrio-EH1       -----MACRTRNLLG---AALFVLLTSAFLLTEANPI--------------GVCIRNCAQCKKMFGPYFEGQL
Tribolium-EH1      -----MDSGSRN----FLVLLLLFA-SSLLVVDANPI--------------GVCIRNCAQCKKMFGPYFEGQL

Pogonus-EH2        -----MVNFQWK-KI--FFSVLIILLTNYICTC--------------SSSVAVCITNCVQCKQMFGQYFEGRA
Ignelater-EH2      ---------MNK-FC--GFLCLLILLWERSVVC--------------NGSLGVCITNCVQCKKMFGPYFQGKA
Photinus-EH2       MTNKDVGPGYNR-LY--GVLYVLVLLWKVPAMG--------------HCSFDICITNCVQCKKMFGPYFQGRV
Aquatica-EH2       --NCAVYFPIMN-VN--VFLCILVLVWKSSTVC--------------YCAFDICITNCVQCKKMFGPYFKARA
Nicrophorus-EH2    -----------M-QT--ILRSMLLIMWLSAYAV--------------EATIPVCLTNCVQCKQMFGRYFQGRA
Aleochara-EH2      -------------MT--SFISTLLLLQLVRVAS--------------TESIPVCLSNCIQCKKMFGRYFQGRI
Oryctes-EH-2       ---------E-K-FG--LYTCYLIFMLLFADFC--------------NASIPVCITNCVQCKQMFGVYFEGKA
Hypothenemus-EH2   ----------MR-IH--LGTLCLLVILMGKHFA--------------KSSIGICLTNCVQCKQMYGPHFEGKI
Anoplophora-EH2    ----------------------MIIVAIKEDLA--------------NASLAVCITNCVQCKQMFGPYFQGRA
Leptinotarsa-EH2   --------MK-C-LH--VFTLSIILVIIEKDLV--------------NASVEVCITNCVQCKQMFGPYFQGKA
Aethina-EH2        --------------------MVIILISNEYYLI--------------TASIPVCITNCVQCKKMYGPYFQGKI
Hycleus-EH2*       --------------------------------------------------------NCVQCKQMYGRYFQGRA
Tenebrio-EH2       ----------------------------------------------------TCLLNCVQCKQMFGPYFQGKA
Tribolium-EH2      -----------------MLKLLFILLAVDTFFS--------------NASIPVCITNCVQCKQMFGPYFQGRA

                  74
Pogonus-EH1        CADACVKFKGKLIPDCEDLSSISAFLNKFE---
Ignelater-EH1      CADACVKFKGKMIPDCEDITSIAPFLSKFE---
Photinus-EH1       CADACVKFKGKMIPDCEDITSIAPFLSKFD---
Aquatica-EH1       CADACVKFKGKMIPDCEDITSIAPFLNKFD---
Nicrophorus-EH1    CADACVKFKGKIVPDCEDIVSIAPFLTKFE---
Aleochara-EH1      CADACVKFKGKIIPDCEDIGSIAPFLTRFD---
Oryctes-EH-1       CADSCVKYKGSIIPNCEDIASIAPFLILNDDLS
Coccinella-EH      CGDACIKFKGKIIPDCDDIASVAPFLTRLD---
Harmonia-EH1       CGDACIKFKGKIIPDCDDISSVAPFLNKLD---
Dendroctonus-EH1   CADACVKFKGKIIPDCEDFASIAPFLSKFDNY-
Hypothenemus-EH1   CADACVKFKGKMIPDCEDIASIAPFLSKFDHY-
Anoplophora-EH1    CADACVKFKGKIIPDCEDIASIAPFLNKFD---
Leptinotarsa-EH1   CADTCVKFKGKIIPDCEDLASIVPFLNKFE---
Aethina-EH1        CADACVKFKGKIIPDCEDIASIAPFLSKFD---
Aethina-EH1        CADACVKFKGKIIPDCEDIASIAPFLSKFD---
Hycleus-EH1        CADACVKFKGKIIPDCEDIGSIAPFLSKFE---
Tenebrio-EH1       CADACVKFKGKIIPDCEDIASIAPFLNKFE---
Tribolium-EH1      CADACVKFKGKIIPDCEDITSIAPFLNKFE---

Pogonus-EH2        CGDACMATGGLVTPDCNNPLTLRSFLKRLY---
Ignelater-EH2      CGDACVLSKGLIKLDCNNPDTFLSFLKRFNY--
Photinus-EH2       CADACVSTNGKIILDCNIPETVVTFLKRFH---
Aquatica-EH2       CADACVQSDGLINLDCNIPETIVTFLKRLY---
Nicrophorus-EH2    CGEACISSNGRDMPDCNNPGTLGNYLKRLY---
Aleochara-EH2      CGNDCIISNGRAAPDCNNPGTIGHYLKRFKIYM
Oryctes-EH-2       CGDACISSNGQISPDCNNPGTLGIFLKRFH---
Hypothenemus-EH2   CGDTCVASYGRLIPDCNNPGTFNGYLKRLYRK-
Anoplophora-EH2    CGDACLASNGRVTPDCNNPGTLGSFLKRLY---
Leptinotarsa-EH2   CGDACLSNNGQLVPDCNNPGTLGNFLKRLY---
Aethina-EH2        CGEACIESDGRLTPDCNNPGTLGTYLKRLY---
Hycleus-EH2*       CGDACISSNGRVTPDCNNPITFGNFLKRYY---
Tenebrio-EH2       CGDACITSNGNLMPDCNNAGTLGNFLKRLY---
Tribolium-EH2      CGDACVSTNGRLVPDCNNAGTLGNFLKRLY---
```

Figure S8. Alignment of Coleoptera eclosion hormone sequences. The asterisk for the second *Hycleus* eclosion hormone indicates the conceptual translation of a pseudogene.

```
                    1
Pogonus             MTGKSTLVCVCATLCVAILVAEMVTAAPTYA-DYE-NSIRDLYELMLQKEA---LEERL---G-HQIVRKSSR
Ignelater           MQGYSALKCFCATLCVIIVVAEMVSAAPTYA-DYE-NSIRDLYELLLQKDA---LEERL---GAHQVVRKSSR
Nicrophorus         ---MLNIKYCCLFLLVLIVATEFTSAAPAYN-DYE-NSIRDLYELLLQKEA---IEERI---GAHQVARKSGR
Aleochara           MQGYPRMKIVCAVLCLIVVMAEVATAAPSYA-DYE-SSIRDLYELLMQKEA---LEERL---GAHQMVRKASR
Oryctes             MQKYSALRCLCAVLCVMVIVADMATAVPTYA-DYE-NSIRDLYELLLQKEA---LEDRL---GAHQVTRKSNR
Coccinella          MQFPVKMRYFFSISCIFLILAALANTAPAYG-DID-NNIRDLLDALYQRDM---LYDRF--NGVHQVVRKAGR
Harmonia            MCSQLKMKYICSIACIFFILAALANSAPAYG-DMN-NNIRELLDALYQRDM---LYDRF--NGIHQVERKAGR
Dendroctonus        MNINNAMKVFSLFVALMLFIASATSA-PSYSGEYE-SSIRELIEMLIQREKENGLEDSR--VGYHTVSRKAGR
Hypothenemus        MNLNSLMKVFAFLASLMLFMACVTAAPPSYPSEYDGASIRELVELLMQRERESGLDDTR--AAYHTVSRKAGR
Anoplophora         MNPSSAIKFFCVIFSILIALAVVSAA--PYSSDYD-NNIRDLFEILMQRDN---LEDRM---GLHQVVRKS-R
Anoplophora-s.v.    MNPSSAIKFFCVIFSILIALAVVSAA--PYSSDYD-NNIRDLFEILMQRDN---LEDRM---GLHQVVRKS-R
Leptinotarsa        MHTNTAVKFFCVIFSILITIAAVSPA--PYS-DYD-NINDLYELLLQQEK---LNDE-L---GRHQVERRA-R
Aethina             MQSVTMKKYLLAVVCIAVLAATISSAAPSYS-DYD-NNIRDLFEILMQRDG---VEDGR--LGLHQVVRKQSR
Hycleus             MQGVAIMKCLCAVTCIMVLFATVSTAPSSYA-DYD-SNIRDLWEMLLQREA---LDEKY--GGGHQLVRKSGR
Tenebrio            MQGYSAMKCLCAVTCIMIVVATVTSAAPSYA-DYD-NNIRDLWEMLLQKDA---LEDKF---GGHQMVRKSSR
Tribolium           MQRYSAMKCLCAVTCIMIVVATVTSAAPSYA-DYD-NNIRDLWEILLQKEA---MDDKFAPGGPHQMVRKSGR

                   74
Pogonus             SPSLRLRFGRRSDPSLLAG-------------------------PPFMLPQSGEPERLEN-----
Ignelater           SPSLRLRFGRRSDPSLLPA-------------------------SQYILAQEAEHGDN-------
Nicrophorus         SPSLRLRFGRRSDPSLIQA-------------------------SPYMLSAAQDAAEIAN-----
Aleochara           SPSLRLRFGRRSDPSLLPG-------------------------GPFMYQAAAAQEAAERAAGDN
Oryctes             TPSLRLRFGRRSDPNMLAS-------------------------PYLLAQESTDHSDN-------
Coccinella          SPSLRLRFGRRSDPELSPG-------------------------EAFLMAQQAEEN--------
Harmonia            SPSLRLRFGRRSDPELSPG-------------------------VAFLMAQQPDDN--------
Dendroctonus        SPQLRLRFGKRMDSQFPVP-------------------------AAYITGLNDITNDN-------
Hypothenemus        SPQLRLRFGKRLDTQYAVP-------------------------SAYMAALNDIPTEK-------
Anoplophora         GPQLRLRFGKRPDAGYEP-------------------------ISAYMSGISADGEN--------
Anoplophora-s.v.    GPQLRLRFGKRPDAGYEPV-----VKRPRSLRLRFGKRSDDLPISAYMSGISADGEN--------
Leptinotarsa        GPQLRLRFGKRAGS-FDPVMKPIPVERAPSLRLRFGKRSEESPISDYLSGLSSEGAN--------
Aethina             SPQLRLRFGKRQDPSLLPV-------------------------SAYMMGEQDN----------
Hycleus             SPSLRLRFGRRSDPSMTPE-------------------------AAFMMAQAIDHNNN-------
Tenebrio            SPSLRLRFGRRSDSSMSPE-------------------------AAFMMAQAVDHDN--------
Tribolium           SPSLRLRFGRRSDASMTPE-------------------------AAFMMAQAVDHETN-------
```

Figure S9. Alignment of Coleoptera sNPF precursors. Note that these sequences are well conserved. The apparent exceptions of *Leptinotarsa* and *Anoplophora* are due to the appearance of an extra exon. *Anoplophora* s.v., *Anoplophora* splice variant.

```
              1
Pogonus       MAATHTLIA---IFCFCMI----ASLVIETQARYLPTRS-YEDRLDKLKEVLRELLYPEE
Ignelater     MSAQKILIT----ALCVI-MVTFLFNTCVVEGRYLPTRS-NTDRLDKLKELLKELLESEI
Photinus      MTAQKILAF----ALLAA-LLAILMSGNVAEGRYLPTRG-NSDRLDKLKELLKELLENQI
Aquatica      MSAQKILTI----TLFAI-MFTFLMNTCFVEGRYLPTRS-NTDRLDKLKELLKELLENQI
Aleochara     MASLKLTSPTTVLMIAGIVLVVWSQQVSDVNARYLPTRGGKMDRLDKLRELLKELLESEI
Nicrophorus   ------MLL----CVLAA--LLLINLSEVSEARYLPTRA-KVDRLDKLRELLKELLESEI
Oryctes       MS-KNVA-----LYIFFALVIMATVLNQEIEARYLPARG-SPERLEKLRELLKELLENEI
Coccinella    MFAKRFGLL----CLLAM--ALVLSFSDTSEARYLSTRA-SGDRIDKLRELLKELLETEI
Harmonia      MFTKHFSLF----CLLVV--VSMLAFSDTSEARYLSTRG-SADRIDKLKELLKELLETEI
Dendroctonus  MTSHAVL------LMATVVVLAALVSEYRVEARYLPTRS-NGDRIDKLRELLKDLLESEI
Hypothenemus  MASQLVLTFRTLLV-----LLIVILAEPQVEARYLPTRS-NTDKIDKLRDLLKDLLESDL
Anoplophora   MFSRSFVIS----ALFVILSAI-LLD-SQAEARYLPTRA-NGERVDKLRELLKELLESEI
Leptinotarsa  MLSKTLFFS----SLFVVFAVL-LLD-ASVEARYLPTRA-NGDRVDKLREMLKELLESEI
Aethina       MFSKQLVLA----MFLVMFAAT-FLDHVAVEARYLPTRS-NGERIDKLKELLRELLENEI
Aethina       MFSKQLVLA----MFLVMFAAT-FLDHVAVEARYLPTRS-NGERIDKLKELLRELLENEI
Hycleus       MFSRRLSIT----VCLVVYII-SVINFYQVDGRYLPTRG-NGDRIDKLRELLKDLLESEI
Tenebrio-1    MFDRKFVFA----VFLVVFATLALETRHTVEARYLPTRS-NGDRVDKLRELLKDLLQSEI
Tenebrio-2    MFARKFFFT----ACLVLFVSLALETSHTVHARYLPTRS-NGDRVDKLRELLRDVSLRSS
Tribolium     MFDRKLVFA----LVFVVFATLAV------EGRYLPTRS-NGDRIEKLRELLKDLFENEV

             61
Pogonus       EKPPMKDSYYRWLENMENNKNAFPPVRY--------------------------------
Ignelater     EKEEF----------Q------PDGPPRWHPESKLFYKREVPAPTQ--------------
Photinus      EKEEY----------Q------QDGPPRWHPESKLFYKREVPNKP---------------
Aquatica      EKEEY----------Q------QDGPPRWHPESKLFYKREVPNSN---------------
Aleochara     EREEY----------Q------ADAPPRWHPESKLFYKREVQQEIPEFDYSNIPTSTIAN
Nicrophorus   EKEEY----------QA-----QDAPPRWHPEQKLFYKREIPQ-----------------
Oryctes       EKEDF----------P-------DQGPRWHPESKLFYKREVPAN----------------
Coccinella    EKED-----------Q------GDAPPRWHPESKLFYKREAPQTKH--------------
Harmonia      EKED-----------Q------GDAPPRWHPESKLFYKREVAETKH--------------
Dendroctonus  DKEES-------------------ETQKWRPDIKYFVKRDVEPRSNEMKSH---------
Hypothenemus  DDTDA----------QRWRSNLETGTLRWNPNSKYYVKREARINNSKSDSVRGKEN----
Anoplophora   EKEEM------------------GDVPRWHPESKLFYKREAPEVQTQPLPQQ--------
Leptinotarsa  EKEEM------------------GDVPRWHPESKLFYKREAPTPKADTQ-----------
Aethina       EKDEL------------------GDVPRWHPESRTFYKREAKPAN---------------
Aethina       EKDEL------------------GDVPRWHPESRTFYKREAKPAN---------------
Hycleus       EKEEY----------Q------ADAPPRWHPESKLFYKREAIQQH---------------
Tenebrio-1    EKEEY----------Q------ADAPQRWHPENKLFYKREAPAH----------------
Tenebrio-2    RDISI---------------------------VLFLSVAGKGTG----------------
Tribolium     EKEEY----------Q------ADAPPRWHPESKLFYKREAPAH----------------
```

Figure S10. Alignment of proctolin precursors.

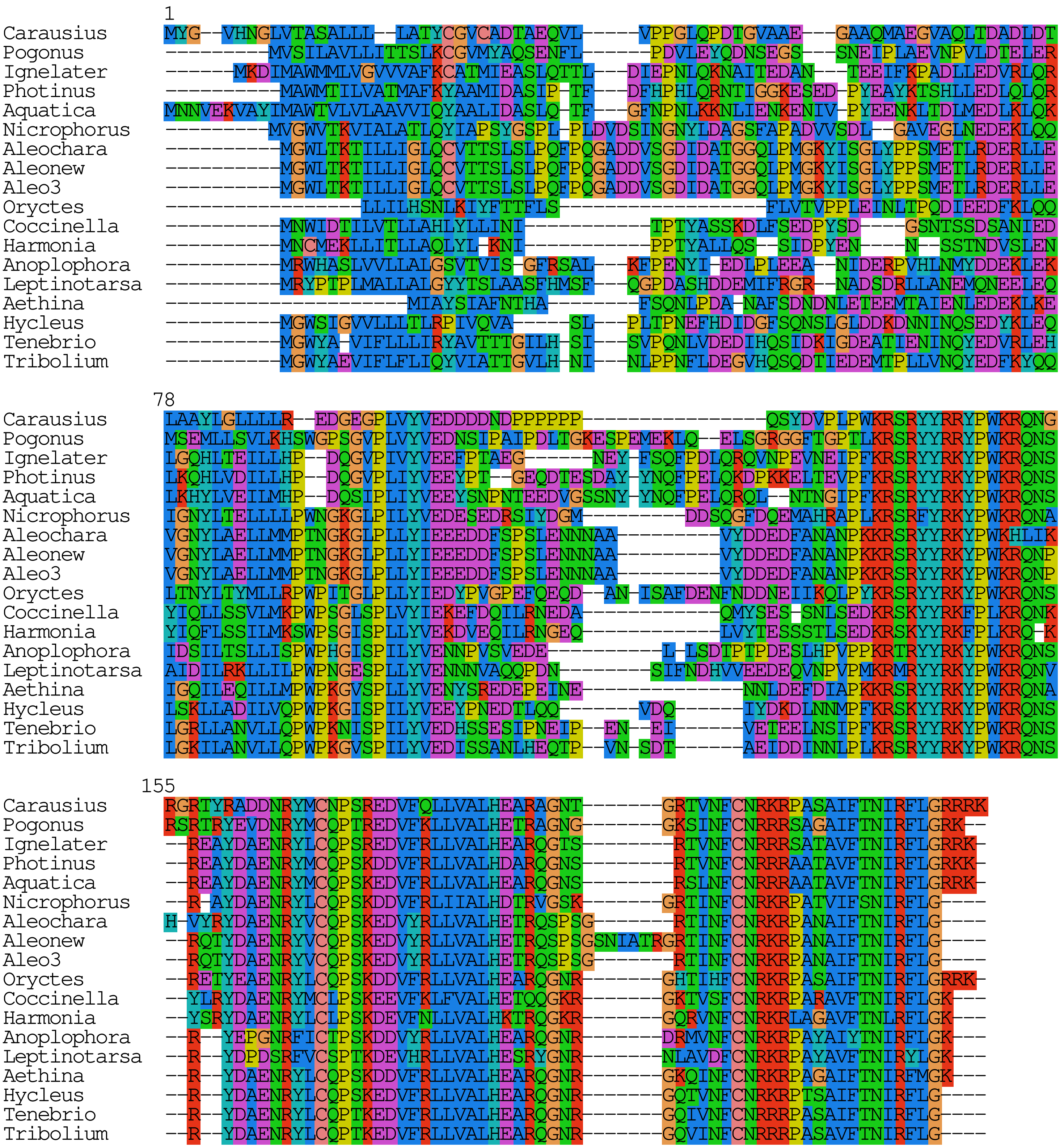


Figure S11. Alignment of RFLamide precursors.

```
               1                                                              *     *
Oryctes-2      -MK---YITLLIFIYFFIYLWRTALIILMLLEGRSLDLLYHN----AIAEASSPTNECRLTPAIR
Oryctes-1      MFKLLFKSVNIEVLLVTLLAWKIAIIFISLDTICTDSKFKS----LQVQAVGSLDECQLTPVIH
Aethina        -------MLRQILDIVPQRLWQITTIFIILGNMCMDPRLSRG--TLEVSGTSTTDECQVTPVIH
Leptinotarsa   -------MFSRSCVWVISKLWYVTLVFIILGNMCMDPRLKTA--TSQVEAASTTDECQVTPVIH
Aquatica       -------MFTNIFQI---KTWF-TILLIMISSMCMLPSKN----KMITEAASTADECQVTPVIH
Ignelater      -------MFTQIFQSATQQLWTTTVLLTILANMCFDSRLSSLNNKIAVEGASSADECQVTPVIH
Photinus       -------MFGK-FVQIRMQTWLILVAVTLIANMCMSSSKV----KTRVAAANAADECQVTPVIH
Pogonus        -----MTHLGF-----SYSFEFCATIIFLLASMCMDPRFSKS---SAVDASSSGDECQVTPVIH
Anoplophora    -------MLPQVANLVLTKVWKLGTLFVIIATMSMDPRLTNIK-NLQAEAASSVDECQVTPVIH
Aleochara      -------MIGDFQFSKFVCLLQYAFLLIVLSGMCLDPKYAK--------GMISTDECQVTPVIH
Dendroctonus   -----MVVLENVCRLVGSALWNSTLVLIFIASMCMDPRLSAA-PKISAEAASLTDECQVTPVIH
Hypothenemus   -----MVLLVKVSNLVYPKIWNQTLVLVLIASMCMDPRLNSN-SK-PVEAASVTDECQVTPVIH
Nicrophorus    -------MYGNILFKVKSFMEKSLIVLIIIGNMCMDPHFQG---KTLVQAANAADECQVTPVIH
Coccinella     -------MIIAAIIHLSCNILTKSIMFMIIGSMCIDPRYIQ----YEVSAASKTDLCEVTPVIH
Harmonia       ------------------NIFCKAIMFLLIGSMCIDPRYIK----YEVAAASKTDLCEVTPVIH
Hycleus        --------MNSPTRCLFQNLWKIVLLL-IINGTCIDPRLN------KVSAASSTDECQVTPVIH
Tenebrio       --------MKSLITLVPWKLWRLTLLFAVLSSMCLDPRLTSN---IKATAASTTDECQVTPVIH
Tribolium      --------MKPLLSLV--TLWKLTI---FLSSMCLDPRLNSK---IQVSGASTTDECQVTPVIH

             65*** *  **** ** *     *             *                   * * *  ****
Oryctes-2      RIKYEGCSSNLIHTFGCTGRCSSYLQVSGSKIWQLIRSCMCCQESGEREASVIINCINPVSEYK
Oryctes-1      VLQYPGCIPKPIPSFACMGRCASYLQVSGSKIWQMERSCMCCQESGEREASVSIFCPQAKPGEK
Aethina        VLQYPGCVPKPIPSFACIGRCASYIQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Leptinotarsa   VLQYPGCVPKPIPSFACIGRCASYIQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Aquatica       VLQYPGCVPKPIPSFACTGQCASYIQVSGSKIWQVERSCMCCQESGEREASVSLFCPKAKPGER
Ignelater      VLQYPGCVPKPIPSFACTGRCASYIQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Photinus       VLQYPGCVPKPIPSFACTGRCASYIQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Pogonus        VLQYPGCVPKPIPSFACIGRCASYLQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Anoplophora    VLQYPGCVPKPIPSFACIGRCASYLQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Aleochara      VLQYPGCVPKPIPSFACIGRCASYLQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGEK
Dendroctonus   VLQYPGCVPKPIPSFACTGRCASYIQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Hypothenemus   VLQYPGCVPKPIPSFACTGRCASYLQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Nicrophorus    VLQYPGCVPKPIPSFACIGRCASYLQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGEK
Coccinella     VLQYPGCVPKPIPSFACIGRCGSYLQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Harmonia       VLQYPGCVPKPIPSFACIGRCGSYIQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Hycleus        VLQYPGCVPKPIPSFACTGRCASYIQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Tenebrio       VLQYPGCVPKPIPSFACIGRCASYIQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER
Tribolium      VLQYPGCVPKPIPSFACIGRCASYIQVSGSKIWQMERSCMCCQESGEREASVSLFCPKAKPGER

            129 *   *    *       *  * *           * *      *
Oryctes-2      RFQKVTIKAPLSCMCRPCNDVEKHNVIPQELAGYSEKSRLSGHFLL----
Oryctes-1      KFRKVTTKAPLDCMCRPCTGIEESAVIPQEIAGYTDEGPLSNHFLKSTTQ
Aethina        KFIKVTTKAPLECMCRPCTGVEESAIIPQEIAGYADEGPLSNHFLKSHQQ
Leptinotarsa   KFIKVTTKAPLECMCRPCTGVEESAVIPQEIAGYSDETPLSGHFLKSHQL
Aquatica       KFKKVTTKAPLECMCRPCTGVEESAVIPQEIAGYTDEGQFSNHFIKSQTQ
Ignelater      KFKKVTTKAPLECMCRPCTGVEESAVIPQELVGYADEGPLSNHFLKSQTQ
Photinus       KFRKVTTKAPLECMCRPCTGVEESAVIPQEIAGYADEGPLSNHFLKSQSQ
Pogonus        KFRKVSTKAPLECMCRPCTGVEESAVIPQEIANYGEESPLSHHFRKSQNQ
Anoplophora    KFIKVTTKAPLECMCRPCTGVEESAVIPQEMAGYSDE-ALSGHFLKSHQL
Aleochara      KFRKVSTKAPLECMCRPCTGVEESAVIPQEIAGYTDEGPLSNHFLKSHVQ
Dendroctonus   KFIKVTTKAPLDCMCRPCTGIEESAIIPQEIAGYTDEGPLSGHFMRSHVQ
Hypothenemus   KFIKVTTKAPLDCMCRPCTGVEESAVIPQEIAGYTDEGPLSGHFMRSHIQ
Nicrophorus    KFRKVTTKAPLECMCRPCTGVEESAIIPQEIAGYADEGPLSNHFLKSHAH
Coccinella     KFLKVTTKAPLNCMCRPCTSIEESAVIPQEIAGYADEGPLSDHFLKSQPQ
Harmonia       KFIKVTTKAPLSCMCRPCTSIEESAVIPQEIAGYADEGPLSDHFLKSTPQ
Hycleus        KFIKVTTKAPLECMCRPCTGVEESAIIPQEIAGYADEGPLSNHFLKSHSQ
Tenebrio       KFIKVTTKAPLECMCRPCTGVEESAVIPQEIAGYADEGPLSNHFLKSHSQ
Tribolium      KFIKVTTKAPLECMCRPCTGVEESAVIPQEIAGYADEGPLSNHFLKSHSQ
```

Figure S12. Sequence comparison of Coleoptera bursicon A sequences. Note that the first sequence, that of the second Oryctes bursicon A has a large number of amino acid residues that are different from the consensus sequence; those are indicated by the asteriks on top of the sequences.

```
              1
Pogonus       --MARIL-WQLILVLV---SILFIVRGQ-LSGDETCETLPSEIHLIKEEYDELGRLQRTCNGEV
Ignelater     MMGFISIFLTTLIAIT---THYSVQCLN-ELGEENCETLPSEINIIKEEYDELGRLQRTCHGEI
Photinus      -MDAVAIFSKLFLTII---TISRIHTMN-DFGEETCETLPSEINIIKEEYDELGRLQRTCHGEI
Aquatica      -MGFAHNFSKLFLTII---TILHVHSLS-DISEENCETLPSEINIIKEEYDELGRLQRTCHGEI
Nicrophorus   -MGIA-RLFFCCF--ILNLYIYTVNSLNEIPGEENCETLPSDIHLIKEEYDELGRLQRTCNGEV
Aleochara     -MATS-RVHTLCLRLLFAILFASIAALNEIPGEENCETLPSEIHLTKEEYDELGRLQRTCNGEV
Oryctes       -MGNTRTYFNVLVIVI--ISLYKANGYSNIPGEETCETLQSEINLLKEEFDELGRLQRSCSGEI
Coccinella    -MALACTRIGLSFFLLY--TIFTLSYCKPDMPEETCETIPAEIHLVKEEYDELGHLQRTCSGDV
Harmonia      -----MAVVRFDIFLLF--AILCVGF-GKEMPEETCETIPNELHLVKEEYDELGHLQRTCSGDV
Dendroctonus  ----MFTKGLLLIFAI---FASYGYCVS-ELPDESCETLPSDIHITKEEFDELGRLQRTCNGEV
Hypothenemus  ----MSSKEFLVFLVL---ISTYVYCVS-ELTEETCETLPSEIHITKEEFDELGRLQRTCNGEI
Anoplophora   ------MLYTFLVSLL---CFSYIYCVS-DMSDETCETLPSEIHLTKEEYDELGRLQRTCNGEV
Leptinotarsa  -----MLFYTVFFGAM---AVSFVYCLN-DMSEETCETLPSEIHLTKEEYDELGRLQRTCNGEV
Aethina       --MAITVFHRIVFACM---MFYYVCCVS-DLAEETCETIPSDLHLTKEEYDELGRLQRTCNGEV
Hycleus       -----MYNYSVFFLIL---YCTYVSSLS-DLSEETCETLPSDVNLIKEEYDELGRLQRTCNGEI
Tenebrio      -----MYN-KILLCLV---YVSYVSSVS-EISEETCETLMSDINLIKEEFDELGRLQRICNGEV
Tribolium     -----MFD-KIILCLV---YATCVYSVS-EISEETCETLMSDINLIKEEFDELGRLQRICNGEV

             65
Pogonus       AVNKCEGACNSQVQPSVITPTGFLKECYCCRESFLRERTVTLTHCYDPDGMRLTNEDMHSMDIK
Ignelater     AVNKCEGTCNSQVQPSVITPTGFLKECYCCRESFLRERVVTLSHCYDPDGMRLTNEDMHSMDIK
Photinus      AVNKCEGSCNSQVQPSVITPTGFLKTCYCCRESFLRERVVTLSHCYDPDGMRLNAEELKAMDIK
Aquatica      AVNKCEGMCNSQVQPSMITPTGFLKECYCCKESFLRERVVTLPHCYDPDGLRLMTADMHTMDIK
Nicrophorus   AVNKCEGTCNSQVQPSVITPTGFLKECYCCRESYLRERIITLTHCYDPDGIRLIGEGKQTMDIK
Aleochara     AVNKCEGTCNSQVQPSVITPTGFLKECYCCRESYLRERTITLTHCYDPDGMRLTSDAMKQMDIK
Oryctes       AVNKCEGSCNSQVQPSVITPTGFLKDCYCCRESYLKERVITLTHCYNPDGIRLTENDMQTMDIK
Coccinella    AVNKCEGSCRSQVQPSVITPTGFLKECYCCRESFMRERIITLNHCYDIDGVKLIDEGNSSLDVK
Harmonia      AVNKCEGSCRSQVQPSVITPTGFLKECYCCRESFMRERIITLNHCYDIDGVKLVEEGTSSLDVK
Dendroctonus  AVNKCEGSCKSQVQPSVITPTGFLKECYCCRESYLRERIVTLTHCYDPDGLRLTGEGENSMDIK
Hypothenemus  SVNKCEGSCKSQVQPSVITPTGFLKECYCCRESYLRERIITLTHCYDPDGVRLAAEGENSMDVK
Anoplophora   AVNKCEGSCKSQVQPSVITPTGFLKECYCCRESFLRERIITLSHCYDPDGVRLTNEGTNSMDVK
Leptinotarsa  AVNKCEGSCKSQVQPSVITPTGFLKECYCCRESFLRERIITLSHCYDPDGVRLTGEGDNAMDIK
Aethina       AVNKCEGSCKSQVQPSVITPTGFLKECYCCRESYLRERIITLTHCYDPDGVRLTGEHNNAMDIK
Hycleus       AVNKCEGSCKSQVQPSVITPTGFLKECYCCRESFLRERIITLSHCYDPDGVRLTAAGVNSMDVK
Tenebrio      AVNKCEGSCKSQVQPSVITPTGFLKECYCCRESFLRERTITLTHCYDPDGVRLTTENANSMDVK
Tribolium     AVNKCEGSCKSQVQPSVITPTGFLKECYCCRESFLRERTITLTHCYDPDGVRLTAETVNSMDVK

            129
Pogonus       LREPSDCKCFKCGDFSR
Ignelater     LKEPSDCKCFKCGDFSR
Photinus      LREPSDCKCFKCGDFSR
Aquatica      LREPSDCKCFKCGDFSR
Nicrophorus   LKEPAECKCYKCGDFSR
Aleochara     LKEPAECKCYKCGDFSR
Oryctes       LREPIECKCSKCGDFSR
Coccinella    LKEPAECKCYKCGDLGR
Harmonia      LKEPAECKCFKCGDLGR
Dendroctonus  LREPSECKCYKCGDFSR
Hypothenemus  LREPSECKCYKCGDFSR
Anoplophora   LREPSDCKCYKCGDFSR
Leptinotarsa  LREPSECKCFKCGDFSR
Aethina       LREPTECKCYKCGDFSR
Hycleus       LREPAECKCYKCGDFSR
Tenebrio      LREPAECKCYKCGDFSR
Tribolium     LREPAECKCYKCGDFSR
```

Figure S13. Sequence comparison of Coleoptera bursicon B sequences.

```
                  1
Pogonus           -----------MR--RPEEVRIALVTLSIMLAFAALHTCQAKRGCSAFGHSCFGGHGKRMDP
Photinus          -----------MYCTN--GSFVAFFAMAALILVIKTDAVDAKRGCATFGHSCYGGMGKRMSP
Ignelater         -----------MNWSN--AAFAVFFAVIALL--LNSEGVEAKRGCATFGHSCYGGMGKRMSS
Aquatica          -----------MNWSK--GAFVSFFAMVALILVIKSEGIEAKRGCATFGHSCYGGMGKRMSP
Nicrophorus       -----------MNSCS--LKMITV-IVSALVLALHYQETEAKRGCANFGHSCYGGMGKRDGG
Aleochara         MSRKTQPVVTIGKSYSGGGFLFGLLLVFLLVFATQSAVVEAKRGCAKFGHSCYGGMGKRANS
Coccinella        ----------MGYS-R------VFPAVLFILLVLNVADSEGKRGCANFGHSCYGGMGKRAPK
Harmonia          ----------MGCCWY------NLLAFLFIFLVLNFGVSEGKRGCANFGHSCYGGMGKRGHK
Dendroctonus      -----------MAIFK---AILVFTLVLLSSLFITFEGVEGKRGCEYFGHSCYGGIGKRTYM
Hypothenemus      -----------MTT----EK--VVTLLFLVIICLVLKRTDARRGCSLYGHSCYGGNGKRANL
Anoplophora       ----------MFTMCR--VTLIL--SAVSLMVLLDVKTTEARSGCVNFGHSCYGGMGKRAS-
Leptinotarsa-A    -----------MSYYT----V-IQLFVITLMLVCSGAEVEGKRGCRSFGHSCYGGIGKRNPG
Leptinotarsa-B    --------------------MFHFVATLAFIL-VLSTITEGRSGCLNYGHSCFGGVAKRHND
Aethina           -------------MSR--SAVLLLPLFMLFVFLLSLDGCDAKRGCANFGHSCYGGMGKRAST
Hycleus           -----------MNCCS--PVGVVVMITTVLIFVFQPDLVEAKRGCATFGHSCYGGMGKRASE
Tenebrio          -----------MNCWS--SVVVLL-AVMAFVLAFHAEAAEAKRGCATFGHSCYGGMGKRASE
Tribolium         -----------MNCWS--TQVVLL----AFVMAFVLAAAEAKRGCATFGHSCYGGMGKRTE-

                 63
Pogonus           SSPIISA-SAPEEVAY--DQQAGLRDVVFTAPRSVSDSAQGPPSLSLLEVAAELQRQHQQDN
Photinus          S-ELMTNDEVLQDVQQE----SNPG-LIFTGPRGDIGSDT----------------------
Ignelater         S-DLLTNEEVLQDVQQE----GNPADIVFTGPRGDVEMEP----------------------
Aquatica          D-DLLTNDEVIQDVQQG----SSPG-MIFTGPRGDIGIES----------------------
Nicrophorus       MM----------EIEDTGKLEENPA-LIFAGSRGDYRPER----------------------
Aleochara         LNEEFTGQELNE------NEQQDPA-FVFTGPRSEYPPLN----------------------
Coccinella        ELMQFT------------DPRQNQG-EIMIGSRFF---NDKPYV------------------
Harmonia          ELLPLL------------DPTQEQG-EILIGSRFI---SDKPYV------------------
Dendroctonus      AE--ASEESMPVEA----QDAEIPA-DVFKGPRSQLPYASREFT------------------
Hypothenemus      MDSGTSKDNSVADL-------ERPL-YVWRESSGSLDNANLPVKSFRL--------------
Anoplophora       -DLIDTNEEVLQN-VE--DVEENPA-FVFTGPRSSYKPKP----------------------
Leptinotarsa-A    LLNASNN---------------------FEKPEYILPSAWGPFRSNQL--------------
Leptinotarsa-B    QNEYSEGEKYFQNDENV-ES--------TDNQRFLYQSEQ----------------------
Aethina           GDLYDTNEEVLQNGFQ--DAEENPA-FVFTGPRSSYQPKP----------------------
Hycleus           --FLDNNEELLQDVQQNEDSVEDPA-FVFTGPRSEFKP------------------------
Tenebrio          --LMENNEEILQDVQGE----ENPA-FVFTGPRSEYKPER----------------------
Tribolium         ----NNNEELLQDVQSE----ENPA-FVFTGPRSEN--------------------------

                125
Pogonus           EKEGRLQRQEAIYGIAPYFRQWLQAYRNSQDFRSQNNM------------------
Photinus          -ERRISLQEL--YNISPFFRQWLQSYRRSQEMHNNGNN------------------
Ignelater         -ERKITLQQL--YNLSPFLRQWLQSYRKLQEMHNGNN-------------------
Aquatica          -ERKISLQEL--YNLSPFFRQWLQSYRKAQEMHNGNN-------------------
Nicrophorus       -EV----------YVSPFMRQLLRNLRTAQQQLQQRDVFDEK--------------
Aleochara         -TQELLRRQEEIYKLPNILGK-----DNDDETEDSGKIFERQTKTLQVDPFLIRQL
Coccinella        -KLKMNRLQY--QQLSRILRQWIHSQTMPQNFEDDKNQI-----------------
Harmonia          -KLKLNPLQY--QQLSRFFKQWISSRS-SQHYEDGQNEI-----------------
Dendroctonus      -QTALSPKQY--EQMNKYIREWVRNYLRKEHELERK--------------------
Hypothenemus      -SKGLADEQQQQQQINKFMKILVSNYLKQLNSEELDN-------------------
Anoplophora       --DRLTQQQY--DNISRVVRQWIANYRRMQEAPNENM-------------------
Leptinotarsa-A    --QIRNPPK--MGQLSAIIAQWISNFKEAQNMPRTSTA------------------
Leptinotarsa-B    -PQIFLEPRY--AEQLRIFRQWLQHHRMAFPAN-----------------------
Aethina           --K-LTPQQY--DNISRIIKSWIQSISKSQERRADNI-------------------
Hycleus           --RRLTPQQY--EHMSRIIRQWIQSYRRAHELQQNNNI------------------
Tenebrio          -PPKLSPQQY--DTISRVIRQWIQSYRGAQEMRENNM-------------------
Tribolium         -QQKLTPEQY--DNISRVIRQWDTIVQESSGNAPRL--------------------
```

Figure S14. Alignment of CCHamide-2 precursors.

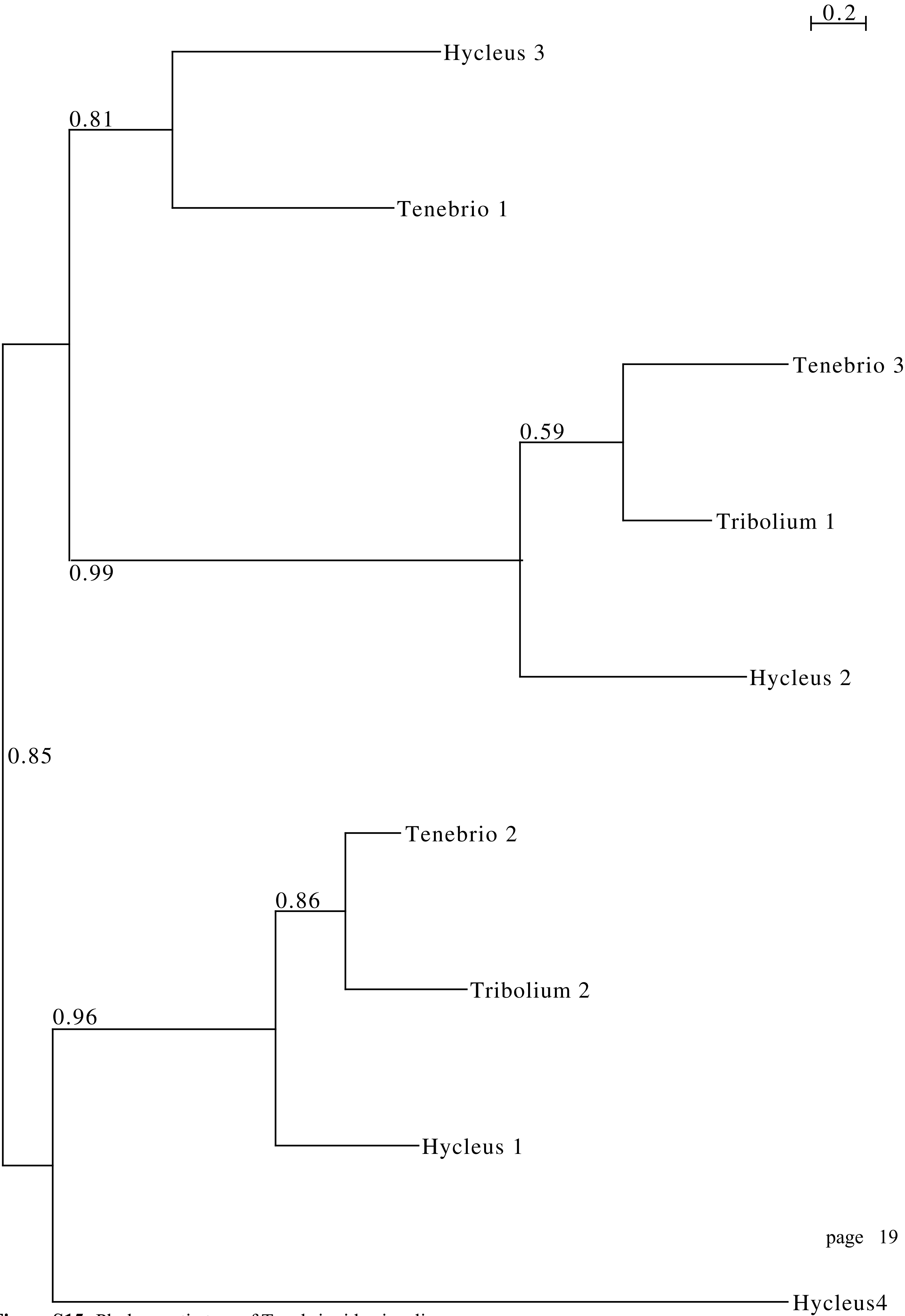


Figure S15. Phylogenetic tree of Tenebrionidea insulins.

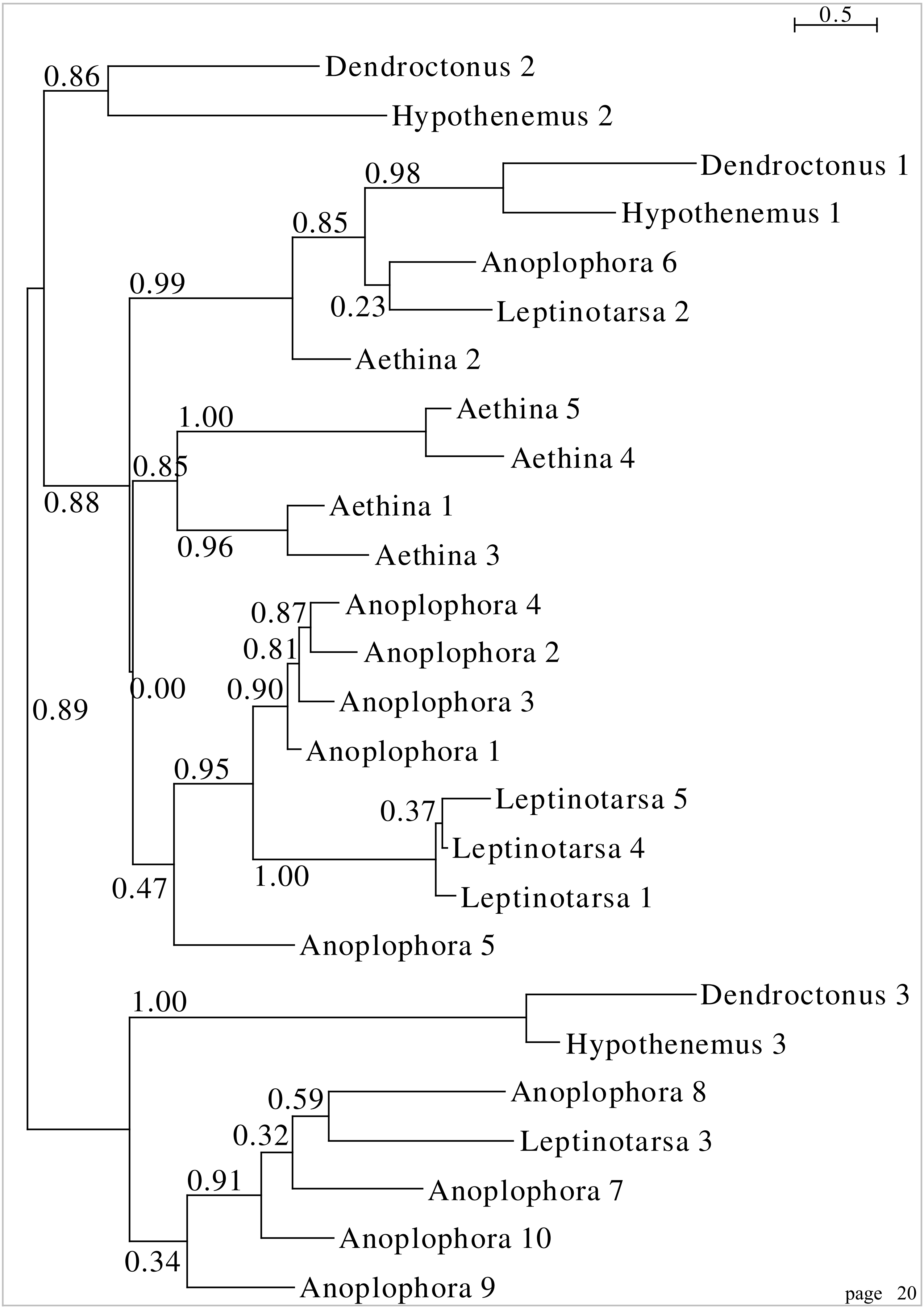


Figure S16. Phylogenetic tree of insulins from *Dendroctonus, Hypothenemus, Anoplophora, Leptinotarsa, Aethina*

```
              1
Pogonus       ----MHSTVLLYAFIG--VVFTTTLCNLGEAHVLSLHCTPCLNDFENGCDDPPA-EICVFGEVR
Ignelater     ----MRATPILPLLF---ALVVTTFSYMSD---ARSLCTPCFT--EEECDKEPE-HFCMWGVAR
Photinus      ----MWVSSIVSLSV-----ILITFTHMTD---ARWVCNPCAT--EDECEREPI-EFCMWGEAR
Aquatica      ----MWGSSLLPLTF---VFIILTFFYSTD---ARWVCNPCVT--EDECEREPI-EFCMWGEAR
Nicrophorus   ----MQTISIISLLFAVT---LSFVITNMAEARAPWICQPCDM--FEECNKEPE-SYCMWGETR
Aleochara     MRSAPPPLSCV-LLVAATICYVSLFGVTSVDARAAWTCQPCVS--AEDCNTEPV-ELCLWGETR
Oryctes-1     ----MDSTMAFIIIGF--ILSSNLV-YL-SSARRFHFCTPCMS--LEDCNQEPN-EVCQWGESR
Oryctes-2     ------------MRKF--IMIVSML-YNTGFGRRNYECRPCMS--MQECNEEPS-DYCPFGETR
Coccinella    ----MKYFSSLLLIIT--IVIL------SDYVQWSAACRWCQG---EECNVAPP-TACPFGEYI
Harmonia      ----MRFFPSILLIVA--IFIL------SDYVRWSTACTWCQG---EECNAPPPQYPCPYGEYT
Dendroctonus  --MNPSTGIILVLCIA-LTLASSIDAMSIT---ERWICTSCVT--QQECDQDPP-SVCIWGEAR
Hypothenemus  ----MHSSLVLMLVFT-LTFIVSVHSMSIT---ESWVCKPCAT--QQECDEKPN-NICVWGEAR
Anoplophora   ----MRSTLVTLLLAV-AIALILLCDLSLA---GKWVCRPCDS--QAECDEDPP-EYCVWGEVR
Leptinotarsa  ----MSSTLVYVVLAL-TVFFILCPGTSIA---GLWICKPCEN--QAECDSQPK-EYCVWGEVR
Aethina       ----MYRFPLVSIILT-VAITLSVFHEASCGSISIKPCRPCI----NDCDDNPP-DECPWGESR
Hycleus       ----MHTSPQILAAILIVVVSFAVFTEQY-NPYRILPCRRCQT--AAECEEEPP-QICMWGEGR
Tenebrio      ----MCPSYNFATIVLVLTITVIIFSDKG-TTM-SLPCRRCFT--SDECNSPPP-DFCPYGENK
Tribolium     ----MCPFHNFITIILVLTVTVIIFSDKG-TAMIHLPCKRCAT--IQECNADPP-QLCVFGENR

             65
Pogonus       DKCGRRVCGKGPGEKCG----GHFGVCGEGMSCTTSFLGANVPRCHGCTLY--ANMECFPVL--
Ignelater     DSCNKKVCAKGPGERCG-GPLGVLGQCGAGMMCYS-D-----EICHGCSS---KTFECYP----
Photinus      DSCNRRVCAKGPGERCG-GPLGILGQCGDGMMCKK-D-----ERCHGCST---QTFQCHP----
Aquatica      DSCNRRVCAKGPGERCG-GPLGILGQCGEGMMCKT-D-----ERCHGCST---QTFQCHP----
Nicrophorus   NSCNRRVCAKGPGERCG-GLRDIFGRCGTGMWCQKED-ESNTYTCHGCYD---NSLECHQSTRY
Aleochara     NFCGRRVCAKGPGERCG-GMKDVLGKCGAGMWCQREEMDSMTFTCHGCYD---NSLDCHRDRRY
Oryctes-1     DKCGRRVCAKGPSERCG-GPMNIYGNCGEGLRCDS-D-----ERCHGCIAVRDSNFVCHPSDDY
Oryctes-2     DMCGRRTCAKGPADRCG-GPNGIYGRCAEGLRCRS-D-----ERCHGCADKGNSDFECYPKF--
Coccinella    NRCRRRACLKGPGEKCGGPTRNSFGDCATGLYCAS-E-----GRCYGCFL---GNLDCYPPQPE
Harmonia      NRCRRRECLKGPGEKCGGPIRNSFGDCATGLFCSS-E-----GRCYGCFL---GNLECYPPQPN
Dendroctonus  NNCGRRICAKGPGERCG-GSLDILGICGEGMMCKS-D-----ERCHGCSM---QTLECSHN---
Hypothenemus  DACGRRICAKGPSERCG-GSLNVLGVCGEGMMCKA-D-----EHCHGCSI---QTMECSHN---
Anoplophora   DACNRRMCAKGPGERCG-GTLNILGQCGEGLMCKS-D-----EICHGCSI---QTMKCYND---
Leptinotarsa  DACNRRACAKGPGERCG-GSLNILGQCGEGMMCKS-D-----EKCHGCSL---QTMQCYNE---
Aethina       DDCGRRVCSKGPGERCG-GYSNAFGSCGEGLTCRPYD-----EKCHGCSL---ITLDCF-----
Hycleus       DACNRRVCLKGPGEKCG-GKFRIMGTCGDGMMCANSD-----SRCHGCYI---PTMTCYPEITD
Tenebrio      NYCGRRVCSKGPGEKCSNDQYAILGTCGEGMWCSNKD-----NRCHGCFI---ATMTCYE----
Tribolium     DYCNRRVCSKGPGEKCG-DRFNILGTCGEGLWCSNKD-----NRCHGCYI---PTMACYPDD--

            129
Pogonus       ------
Ignelater     ------
Photinus      ------
Aquatica      ------
Nicrophorus   ------
Aleochara     A-----
Oryctes-1     ------
Oryctes-2     ------
Coccinella    RKITQY
Harmonia      KNVNQY
Dendroctonus  ------
Hypothenemus  ------
Anoplophora   ------
Leptinotarsa  ------
Aethina       ------
Hycleus       ------
Tenebrio      ------
Tribolium     ------
```

Figure S17. Alignment of Neuroparsin precursors.

```
Pogonus           ----MNNQQ-------VISSLVFVVLIVGINAGLEKLD---SNT-KISYDM---------VGHRL---KR--ST-
Ignelater-1       ----MPSYRLECCAAFIVISCVITAAIAEDHNENDKLA---NTAIPQKYQWYV--------SPRLGRKKKHVSPF
Ignelater-2       ----MGRSGLECCAVFLIVSCVFALIIAEEHNGNDQLT---NTAVSRDLQWYVRRKPGVWLTPRVGRKKRSASPY
Ignelater-3       MSTSSRLYKNVLTFRSVHKNHDIIVAIAEDHNENDKLA---NTATPQKYQWYV--------SPRLGRKKKNVSPL
Photinus-1        ----MGQFCLKCCVLLT---FLVVVTIANQYNGNHHI--------LMAVQLYSNKKPTVLLSSNAQRQKRN-SQY
Photinus-2        ----MKQPCATCTVLVAIVI----MAVSQVHTEVS----------------------------------------
Photinus-3        ----MKQFYATCAILLTVVS----MAIA-----------------------------------------------
Aquatica          ----MGQSSLKCCAVIFVLNLLFAITTADQSNGDHRK--------VIGLQWYVNKKPNTVLSSHILRKKRN-SNY
Nicrophorus-1     ---------MVRQAIFVLAVLILV---AICHSA-------D-NDERSIDSYSKRKMSALWFGPRLGRKKRNPSS-
Nicrophorus-2     ----MHRQVIVLSVLLVSALLISA-----A---------------------------------------------
Nicrophorus-3     ----MHRQAIVLSVLLVSALLTSA-----A---------------------------------------------
Oryctes-1         ----MEHLPVVSCVLLLLCSICVFTEQA----------------NQADTRLKLHTASPFWFGPRLGRNKRNPFS-
Oryctes-2         ----MDRSLYTFCALLLIFYTVIA----D-----------SSNEKTVDEYAKRKQISPLWFGPRLGRSKRNQHF-
Aleochara         ----MDRLVLFICVLLITIQTISS---------S------DQQEATGVDGQTNKKYARIWFEPRMGRRKRHPT--
Harmonia          ----MNRFIWFNCILLLFNTFDGSAVSL-NDYE--------LVEFRKPHHKKHYPAFYSWEGSTMK-EKREPID-
Coccinella        ----MERFVWFNCVLLLLYTIDGSAVSL-NDYE--------LVEFHKPYHKRYYPSFYSWEGANIK-DKAEPIE-
Dendroctonus-1    ----MIRISVLNSFLLLIAVLYYATDSVDS--------------FDPAGDHERKESSPMWFGPRIGRKKRNPKD-
Dendroctonus-2    ----MGFSVTVICVAIFFLILRANPSASIEHGNNINGDPSETEINSRGSSLSQENYSPAWMDRKFGKQKINDLL-
Hypothenemus-1    ----MSNISVFNCFVLLAILLYYTTEIAES--------------LDAPN--DRKTSQAMWFGPRIGRKKRNPTD-
Hypothenemus-2    -------MLRVTCIILVFTTVISMS----STFVL--------SNNIESDATNTAVEKFPWINSRKS------KT-
Anoplophora-1     ----MAKVTLLSFMVFLTLALYFNGVLTA----N------SDELIDAKRRDGEKKYSPLWFGPRIGRRKRNPTE-
Anoplophora-2     ----MDRIIIVACASLIIILNIYSHMVLATHDYDS-------SLYSSKYREIDSKAAPLWFSSNLGRRKRNLKN-
Leptinotarsa-1    ----MSKLSVFTFAAIFIMLYFKSVASA---------------EVDEKRKDEAAKYSPFWFGPRIGRRKRNLID-
Leptinotarsa-2    ----MDRIVLVSCVVILIGNFCFELILASQRFDG--------SVETKMKEEEESKMSPAWFSPRLGRRKRNSKE-
Aethina-1         ----MKQSTKINFALIFMSLIILEICCAESHHGN------LSADKPEHEKHA----SELWHGPKLGRKKRNPSD-
Aethina-2         ----MTRFTLVNCAVLILIYLELAL----CSQPT------NVVAALDKQRDDEVKLDNLWFGPRLGRRKRNDSE-
Aethina-3         ----MRQSTRINCALVFISLIVLEVYCTGSHNYE------NLPDKSGKKTGYEKYISHLWSGARLGRKKRNLDD-
Aethina-4         ----MKQLTRINVALIFMSLVVLEVCCTESQHGN------LSADKSEKKTYH--DKHASQSLPKLGRKKRNPSD-
Aethina-5         ----MSQLAL-TCTILMA--MCLHIVLAAPSN-----------D-VTTDKRNQKTANQMWFGPRLGRRKRTPTP-
Hycleus           ----MDRINLVNCAVLCIIVLYFGIVLAVPHYE-------SAQD-------EGGDKENFWFGPRLGRKKRNQSN-
Tenebrio          ----MERIILVNLAVLCVAILLSEVVLSVPHYG-------SHQVSVRRERNDDKKQSYMWFGPRLGRKKRNSN--
Tribolium         ----MERFILINWTVLCVAVLFFETVLSTPHE-----------SSVPNERNDDSKETYFWFGPRLGRKKRNSSN-

                  76
Pogonus           NEDITPGLYYNKDDLHAIAELIQENP-WILANIPD----------MQK-------RQMYFTPRLGRDLSEENSNS
Ignelater-1       EE-TYELSSLDKEQLESLLGNIQEPPEWTAYTISD----------GKR-------N-SNFVPRLGREIIEIAEPD
Ignelater-2       EENNYRTPSFDREQLESLMDTVQEPP-WTLVTINE----------GKR-------HTVNFTPRLGRESGEENEPD
Ignelater-3       EE-THELSSLNKEQLESLLDNIQEPPGWTAYTISD----------GKP-------N-SNFVPRLGRELIEETDPD
Photinus-1        EDNYRIPAPLDKEQLEALIEAVEEPP-WTVLTFNE----------GKR-------HTVNFTPRLGRESGEEEAE-
Photinus-2        ---KLMNFPADNEQVETMIEATQEHP-WAIFTFGG----------KKR--------SVSFIPRLGRELGDENME-
Photinus-3        ------------------------DE-RNEFFIGG----------KKR--------GVSFIPRLGRELGDDNAE-
Aquatica          DDAFTISSPLEKEQMEAFLKALQEPP-WT-LTFNE----------GKR-------HTVNFTPRLGRESSEEENE-
Nicrophorus-1     DEL-LKNTNLDREQLVALLEMLQESP-WAVVALNE----------GKRH------NKMNFTPRLGRESGEDLAGS
Nicrophorus-2     --------------------FDGQND-YR-SMFN---K-------GKP-------SIYDFVPRLGRESGEDMNAY
Nicrophorus-3     --------------------IDGRND-YQ-SMFT---K-------GKP-------SIYDFVPRLGRDSGEDVNAY
Oryctes-1         PDT-YSLSILTCEDVEE---YVKVIP-WDLVTKCMISE-------GKRDI---KSRMNAYIPRLGRESGEEFISS
Oryctes-2         DDL-EILLASECDEIDEYIRSIPQSM-FERCSINEDKR-------DKP-------KSKQFSPRLGRDSYEDYFGM
Aleochara         MEY-YKTPFMDHQQLETFLEFMQQSP-WADFTLPPNEN-------AKR-------YDVNFVPRLGRESGEEMGNA
Harmonia          DEY-LKFPDLDRK---------SFSS-WLLR-MND--------------------AGRIFPSRINRESNDNELSF
Coccinella        DEY-VKYSDLDRK---------SFSS-WLLR-MND--------------------AGRIFPNRINRESNENELSF
Dendroctonus-1    NEI-FIN--SPDQPTVNLLDMLRETP-MVVLAFSEA----------F--------KQHNFVPRLGRESGESLNGY
Dendroctonus-2    ---------RDKNKMVHILETLDEYL-CSIFTGNDG--------------MQKRDGHLYYTPRLGRNVEVVDNAR
Hypothenemus-1    PKT-YIN--DKNQPSMDLYEALRESP-LVVVAVNEA----------N--------KQHNFTPRLGRESGETLPNW
Hypothenemus-2    DVVWF----TGKIKVSKLLEYLQEYP-WTVIALDSRSG------------IEKRSNSVNFTPRLGRNLDEEGYSA
Anoplophora-1     NIF-KT---LDREDLDA---YALKNP-WPLMTFNN----------GKR--------QMDFTPRLGRDSMEDQEER
Anoplophora-2     SV--------TQHELIDLWEALAKSA-TALVTIDGNDEFVCRSMPNNRFYLVEGVDKSEFNPRLGRKIE-EIPGN
Leptinotarsa-1    ESY--EN--ADNENVEELLDFIRKSP-WTIMLFNH----------GRG-------HIVNYNPKFSKESPDGINEA
Leptinotarsa-2    ETY-R----SSEQELADLLEAFEDSP-LAFIAVNGNEKFICKNIPGKR---------SNFIPRLGRNVEDDFSGS
Aethina-1         DLF-REE--LEQKEQANLFDMLQDTP-WTVVAVGD----------GKR-------HVSSFTPRLGRELQDDFGSN
Aethina-2         DH--YNY--LNAEQIQNILEAIEQSP-YTIVILNP-----------TKR------HTVNFAPRLGRDSGEDD---
Aethina-3         EVF-REE--QKE-QLDNLLQILQDNP-WAIVAMTD----------GKR-------HVSSFTPRLGREVDDDFGSN
Aethina-4         DVF-REE--MEQKEQANLFDMLQDTP-WAVVAVSD----------GKR-------HVSSFTPRLGRELEEDFGSN
Aethina-5         AEGYYDY--LEADEIQSILEAIEGSP-YSLVVMNP-----------NRH------KSLSFAPRLGRESGEES---
Hycleus           DET-YKN--SETEQLATLMDAVQDSP-WAILAF----K-------GKR-------HTLSFTPRLGRESGEELTN-
Tenebrio          IDP-YRN--REREQLATLLDVIQDSP-WAIVAV----N-------GKR-------HVVNFTPRLGRESGEEFVSS
Tribolium         DDL-YQD--MQKEELVSLTDALQDVP-WAIIAVNDLLE-------GKR-------HVVNFTPRLGRESGEEFVNN
```

```
               151
Pogonus           --------DMLN----SGDFKSLRSLQFVPRLGRQLRGHRFY------------------------
Ignelater-1       -------AIWMYDTDLNRELITQRSPPFTPRLGRG-TNKNYHFDPRLGKRSSN-------------
Ignelater-2       --------SWLHDTDLNRELMSQRSPPFAPRLGRR-MN-TYYFDPRLGKRNSNSQKQLL-------
Ignelater-3       -------TIWIYDADLNRELITQRSPPFTPRLGRG-TNNKYHFDPRLGKRNSNLQKQLL-------
Photinus-1        --------NWFRDAELSRELMTQRSPPFAPRLGRR-TH-SFYFDSRLGKRNEQQKM-L--------
Photinus-2        --------KWFRDGEM----KTERSSPFVPRLGRESLIQKPYFVPRLGKREE--------------
Photinus-3        --------NWFGDVEL----RAGR----------AKENQGYYFVPRLGKRAV--------------
Aquatica          --------DWFPDSEVRKELMFQRSPPFAPRLGKR-LN-TYYFDPRLGKRDGRQKM-L--------
Nicrophorus-1     TNPNVGANRWIQDTEINGDLMSQRSPPFAPRLGR----RVVPFSPRLGRENDRLF-----------
Nicrophorus-2     -VINLGDAKWLQDPESSPEIIFQRSA-FSPRLGK-----KSSFSPRLGR-----------------
Nicrophorus-3     -VINLGEGKWMQDPE-VPEVLFQRSA-FSPRLGR-----KSPFSPRLGR-----------------
Oryctes-1         -GPSGVTNKFVDNAEAFREALSSNSPPFAPRLG---------------------------------
Oryctes-2         -----SNDRRPQNSE-----IFVRSPPFAPRLGR----RVLPFTPRLGRETVISY-----------
Aleochara         --------RSNMESELVSDAISQRSPPFAPRLGK----RVLPFAPRLGRETFQLDRA---------
Harmonia          -----TNERWLDID------LASRSPPFAPRLGRR---NVSPFIPRLGRDDKLTFI----------
Coccinella        -----TNERWLDID------LASRSPPFAPRLGRR---NVSPFIPRLGRDDKL-------------
Dendroctonus-1    ----LENNA-DGDVDSLSSK-QRHVNNFPPRLNRN---TYNPYSPRLGRESDLPFK----------
Dendroctonus-2    -------DISLTDG---------SSFSTSRRSRRP---HGSSYSPRLGRDESGIYL----------
Hypothenemus-1    ----ADDAGMVVDGDTLPSR-PNIVNPFAPRLGRN---NYNPFNPRLGRDSEMPLQ----------
Hypothenemus-2    ----G---------------AAEKSF----FYGKG---HGNMYSPRLGRDESTSYA----------
Anoplophora-1     -------------PD----YEEFTHKPLPPRVGR----RYSPYSPRLGRENEILLQ----------
Anoplophora-2     -------EKWLRDIVT--AEVDERSPPFAPRLGR----RLSPYSPRLGREA---------------
Leptinotarsa-1    --------------D--LTEVVERS-MFAPRLGR------SMFAPRLGRAMFAPRLGRAEGEMELE
Leptinotarsa-2    -------EKWLQDSDGSAEYMIQRSPPFAPRLGR----RSSPYSPRLGRDM---------------
Aethina-1         -------------M---EFELSGRSP-FSPRLGK----RMTPFSPRLGR-----------------
Aethina-2         ------NNEWVQESDLLDKNMNERSPPFAPRLGK----RLIPFAQHLSREKYKF------------
Aethina-3         -------------DSMLQFDVAERSPLFSPRLGRS---RSSAFSPRLGRFAGNF------------
Aethina-4         -------------V---EFELSERSP-FSPRLGK----RMTPFSPRLGRYAERF------------
Aethina-5         -------DEWMQVPRSMKKMLHHNKY----------------------------------------
Hycleus           ----PSNDQWGQSPEVYAELLSERSPPFAPRLGR----RLSPYSPRLGKRNDRS------------
Tenebrio          ----AAEDRWLQDPEMSGEMLSQRSPPFAPRLGR----HLSPFSPRLGRENDKNLF----------
Tribolium         ----APEDRWLQNHETSGEMLYQRSPPFAPRLGR----HSSPFSPRLGRENDRNLFS---------
```

Figure S18. Alignment of pyrokinin precursors.

```
              1
Pogonus       MWFPLSTV-AALCMLLDHFDAK-IYDKELEMAFRQRTRSDWERLWHQEKHARCRESIVKHMKWACEKDIYRITRRSD
Ignelater     MWLPVSTV-AAICVIFDAADATISRDNELEDIFRQRTRSDWEEAWHREKYTRCREMLLQHLYWACEKDIYRLTRRSQ
Photinus      MWLPVSTV-VVLCVVLD---GTVSKDNDLENIFRQRSSSDWQEAWHHEKFTRCRDTLIQHLYWACEKDIYRLSRRSE
Aquatica      MWLPVSTV-AVLCVILDLTKATISKDNDLENDFRQRSKSDWQDAWHREKFTRCRDTLIQHLYWACEKDIYRLTRRSE
Nicrophorus   MWLPVSTV-AALCLLFDGVKAR-GMDKEMDLIFRDRSNTEWKEAWHKERYTRCRETLIRHLYWACEKDIYRITRRSE
Aleochara     MWLPQSTLFTTLCLLLVEA----LNNRELEMKFQLRTPGEWEREWHQERYSRCRDSITRHLRWACEKDIYKLKRAEK
Oryctes       MFFPVSTV-AALCVLVKVADGSSASSNELNSIFRGRTKSEWEEAWHREKFVRCRESLMSHLYWACEKDIYRITRRSK
Coccinella    MWLPLPT-FAVLCVLMDVYEST-QVDNELELIFKQRSHSDWREAWHREKYVRCRETLIKHLFWACENDIYRISRRRP
Harmonia      MLVPFLTLAVVFCVLLNVYECT-QVDNELELIFKQRSHSDWREAWHREKYVRCRETLIKHLFWACENDIYRINRRRP
Anoplophora   MWLPVSTI-AAICVLFDACETS-NSNNDLEIAFRERTDSDWKNAWHKEKYSRCRETLIRHLYWACEKDIYRLTRRSD
Leptinotarsa  MWFPVSTI-AAVCVLIDVSDGM-RSENDLELVFKERTHSDWRNAWHQEKHSRCRGDLVKHLFWACEKDIYRLTRRSD
Aethina-1     MKFPVSTI-ATLWLLLDITDGA-MPDRELDLIFRDRSKTDWQNAWHKEKHTRCRERLIGHLYWACEKDIYKLTRRTD
Aethina-2     MWFPVSTI-AAIFVLFDVSDTM-VPDNDLELIFRTRSKSEWQTAWHKEKYSRCRDTLVRHLYWACEKDIYRLTRRTD
Hycleus       MRFPVSTI-ATIFVLFDL---A-SSNNDLELVFRDRSRLDWENVWQNEKHKRCRESLIRHLYWACEKDIYRLTRRSD
Tenebrio      MWFPVSTI-AALCVLLDVSDTT-RSDNELELVFRDRSQSDWEEAWHKEKYTRCRETLIKHLYWACEKDIYRLTRRSD
Tribolium     MWLPVSTI-AALCVLLDMSDTT-RSENELELVFRDRSQSDWEEAWHKEKYTRCRETLIKHLYWACEKDIYRLTRRSD

             78
Pogonus       FT-----------------NIHEMFPWIQETRAKRMIRLRRSGDKR-RSSITNECCSSAGCTWEEYAEYCPTNKRIT
Ignelater     QPKN-VNFF-------DNEKQYLNFPWLTPKQAQQIVRSRR-----GGSSITSECCSSTGCTWEEYAEYCPTNKRHT
Photinus      KPRV-I-KL-------GERYEYDKPPWISVMRARQLVRSRR-----GGSSITSECCSSSGCTWEEYAEYCPTNKRHT
Aquatica      RNNL-K-PY-------KEENNSFKFPWISAMAAKQIIRSRR-----GGSSITSECCSSTGCTWEEYAEYCPTNKRHT
Nicrophorus   HPMQL---A-------NEDLESLSFPWISDEEAHFMVRTKRAANRNGGSSITAECCSRKGCTWEEYAEYCPTNKRYT
Aleochara     PDIYRLKRIENELEDEDVQEYESFYPYYLPETAMAMLRTKRGLSKRSG-IITQECCSPKGCTWEEYAEYCTQNNRKV
Oryctes       PAVQQIDFE-------TGKHMVVDFPWIPKKHALVYLRYRRGYSARSSHSITSECCRSSGCTWEEYAEYCPINKRYV
Coccinella    EKRFN-----------NIFDEDTKYPWIPEKRAKLFIRSRRGINRRTG-SITSECCKSSGCTWEEYAEYCPTNKRYT
Harmonia      AKRSD-----------NIFDEDVKYPWIPEKKAKIFLRSRRGMNRRTG-SITSECCKSSGCTWEEYAEYCPTNKRYT
Anoplophora   PS----YKY--------YNDNELDYPWLVPNKAKRLLRFRRNINRRSGSSITNECCKSSGCTWEEYAEYCPTNKRYT
Leptinotarsa  PRYKSYDDY-------DNDDIEVDFPWKSPRRAKRILRFRRSIDRRSGSSITNECCKSTGCTWEEYAEYCPTNKRYT
Aethina-1     PTFKN---Y-------FMESDELNYPWLIPSKAKSLLQFKRGIK-RSGSSITSECCMSTGCTWEEYAEYCPNNKRHT
Aethina-2     PYFMV-----------VLWRTSDSFPWISPTKAKRYLRFKRDVK-RSGPSITSECCKSTGCTWEEYAEYCPTNKRHN
Hycleus       PTFI---------------NDKEQFPWLSVEGAKSFLRFRRASSRRGGSSITAECCKSTGCTWEEYAEYCPTNKRHT
Tenebrio      QNG--FNNY--------INNVDEEFPWMAPKKAKRLLRFRRGVNRRAGSSITSECCKSSGCTWEEYAEYCPTNKRYT
Tribolium     Q-S--YNNY--------ITNTDEEFPYLAPKKAKRLLRFRRGVNRRAGASITSECCKSSGCTWEEYAEYCPTNKRYT

            155
Pogonus       PFV--------
Ignelater     VYVD-------
Photinus      VFVD-------
Aquatica      VFVN-------
Nicrophorus   NYL--------
Aleochara     PHKRYEYISDY
Oryctes       -----------
Coccinella    SYV--------
Harmonia      SYV--------
Anoplophora   SYI--------
Leptinotarsa  SYV--------
Aethina-1     LYSSY------
Aethina-2     SVLNSYL----
Hycleus       PFVSRF-----
Tenebrio      SYV--------
Tribolium     SYV--------
```

Figure S19. Alignment of relaxin precursors.

Figure S20. Alignment of vasopressin precursors.

```
               1
Pogonus        -MKSKGFEVIY---C--TLFVLSVISMCIARPQQSINDLSPQAVGSGTGGGRLSESAAVRNLI
Ignelater      MSSNQTAQLLARSFL--IIVVVGVVSVS-ARAAHFGDSNQVNPDQE-SAA---------N-L-
Photinus       -MPQQIMQFLMRSIL--LLLVLATVSTSARSTTHFGETNQINQEQDGSSG---------G-L-
Aquatica       -MSNQAAQVFVRFFL--IIALMTTISVS-ARSAHFEEASEVNSEPDGPQA---------N-I-
Nicrophorus-1  -MVCNMVSLISRSVL--VMLIV--SSLVSARPQHFGDSSQGLSAEDLAG------------L-
Nicrophorus-2  -MVCNMVSLISRSVL--VMLIV--SSLVSARPQHFGDSSQVLGEQESDGG---------NAL-
Aleochara-1    ---------MMRNFILLLAVAVASFALVSARPQKYIDSLSQAGDG--VDD--VA------DNQ
Aleochara-2    ---------MMRNFILLLAVAVASFALVSARPQKYIDSLSQSAPL--SALTPLAEADSNTNEV
Oryctes        ----------------------------------------PSSDQDTSNM---------V-D-
Coccinella     -MNTDLVRI----KS--FLLLLIVVGAVAAKPSHFSDNQVTPDAGDRSDI-------------
Harmonia       -MSSDLVRL----TS--FLLLCLLVASIAAKPSHFPDNQVTPD-GDRSDM-------------
Dendroctonus   -MQYEFPQVISRVF---FAILIMSLVAVNAKPPHYQENT--QGLNADQE-------STLNNV-
Hypothenemus   -MQLEFSQVIPRLFL--FAIFFVSLASVMGKPPNFQETASPQVFNNDPD-------PVINNN-
Anoplophora    --MQDLSQFF-SKYV--FILLVVTLIFVNARPSHLSESSQ--LL-P----------DQESNM-
Leptinotarsa   -MMQGFLHLFMKAF---LILLVAALTT--SNARHLGENSLPENSQ-FMV-------DPDNNQ-
Aethina        -MSSEIPVFLT-KAL--LILIVVSVAVANARPSHFGENNQAV----SEQ-------ESNNNI-
Hycleus        -----MSQPNNLGPV--VVFLVALLASTKARPSHFADNQL-LPETD-------------TNL-
Tenebrio       -MSAQVPHYLFRTLL--VL-FVATLAVSSARPNHFGDPNQVVAE-G--DA---------NNL-
Tribolium      -MAAQLPRYLTKTLF--IF-LIATLVVANARPNHFGDASQVVGEPA--DG---------NNL-

              64
Pogonus        DTDDD-LTALK-MQLLA---QQLA-EYNAVP---------QLPDYAA------------DRF-
Ignelater      -VDSG-LTPLQ-LEMLA---QRLS-EYNNQI---------PQVPAAW------------DRL-
Photinus       -IDSG-LTPLQ-LEMLA---QRLS-EYN-QI---------PPIPAAW------------ERL-
Aquatica       -VENG-LTPLQ-LDLLA---QRIS-ELN-QI---------PPIPIAW------------ERL-
Nicrophorus-1  -L----EDPMV-LAYLR---KDVE-RILDAA---------GSSDYEW------------DTM-
Nicrophorus-2  -GSPG-FSDLQ-LEMLA---HRLA-EYNR------------DLPVGW------------ERM-
Aleochara-1    LTDMQ----F---ELIA---QQLAN-----L-N-SRP----QF----------------AAAM
Aleochara-2    LPPMA-LLQLGLWQMLD---DRFDDDFSP-YSH-SRSLRQRQHDGHLKQALLAQSTRSQNGIF
Oryctes        -PSAG-WTNEQ-MEQLL---QHLA-ELN-RV---------PQYPYGW------------ERL-
Coccinella     -MDPGLINAWRRYDFLM---RRLAADYDNGN---------MANDNPL------------DRL-
Harmonia       -IDPGLLNAWRRYDLMM---RRLTADYDNAN---------MANENPL------------ERM-
Dendroctonus   -LDSG-LRPWQ-LELFA---QRMA-ELNQ-LSQMNNNLNGQEY-GTF------------DRAL
Hypothenemus   -LDSV-FRPWQ-LEFIA---QRMA-ELNQ-LGQMNN-LNGQEYGGGF------------ERAL
Anoplophora    -IDSG-YKQWQ-MEMLA---QKLA-ELNQ-LGNGEY---------GL------------ERSL
Leptinotarsa   -YEPG-MKQWQ-MELIA---QKLA-ELNS-P-------------IGF------------QRDL
Aethina        -LDGG-LKPWQ-YEQLLAQAQRLA-ELNG-NDQDA---------FGF------------ERAL
Hycleus        -LETN-FKPWH-LELLA---QRLS-EMNLPAAQQMNNINNNIDDYGW------------ERAL
Tenebrio       -LDSG-LKPWQ-LEMLA---QRLS-EIS--Q---------TGGDYGW------------DKS-
Tribolium      -LDSR-LKPWE-LEMLV---QRLS-EISS-Q---------TGGDFAW------------DKS-

             127
Pogonus        -------VLSKRQQR-FRQCYFNPISCFRK
Ignelater      ----ARNPEVKRQSR-YRQCYFNPISCFRK
Photinus       ----ARNPEVKRQSR-YRQCYFNPISCFRK
Aquatica       ----VRNPEVKRQRR-YQKCYFNPISCFRK
Nicrophorus-1  ----D--MPSKRQTN-RLKCYFNPVSCF--
Nicrophorus-2  ----IRSPEMKRQSR-FRQCYFNPVSCFKK
Aleochara-1    DKHLFRLPEMKRASR-FRQCYFNPVSCFRK
Aleochara-2    QSGLPG-MPVKRQGK-RLKCYFNPVSCF--
Oryctes        ----IRGPEMKRQSR-FRQCYFNPVSCFKK
Coccinella     ----FRNPEMKRQGRSYRQCYFNPISCFKK
Harmonia       ----FRNPEMKRQGRSYRQCYFNPISCFKK
Dendroctonus   RS-G---NEIKRQSR-YRLCYFNPVSCFKK
Hypothenemus   RSER---NEIKRQSR-YRLCYFNPVSCFKK
Anoplophora    RS-G---NEIKRQSR-YRLCYFNPVSCFKK
Leptinotarsa   RS------ETERKRR-FRACYFNPVSCFKK
Aethina        RS-R---SDVKRQSR-YRLCYFNPVSCFKK
Hycleus        RERGIRSPESKRQSR-YRQCYFNPISCFKK
Tenebrio       ----IRSPESKRQSR-YRQCYFNPISCFRK
Tribolium      ----IRLPEAKRQSR-YRQCYFNPISCFRK
```

Figure S21. Alignment of allatostatin CCC precursors.

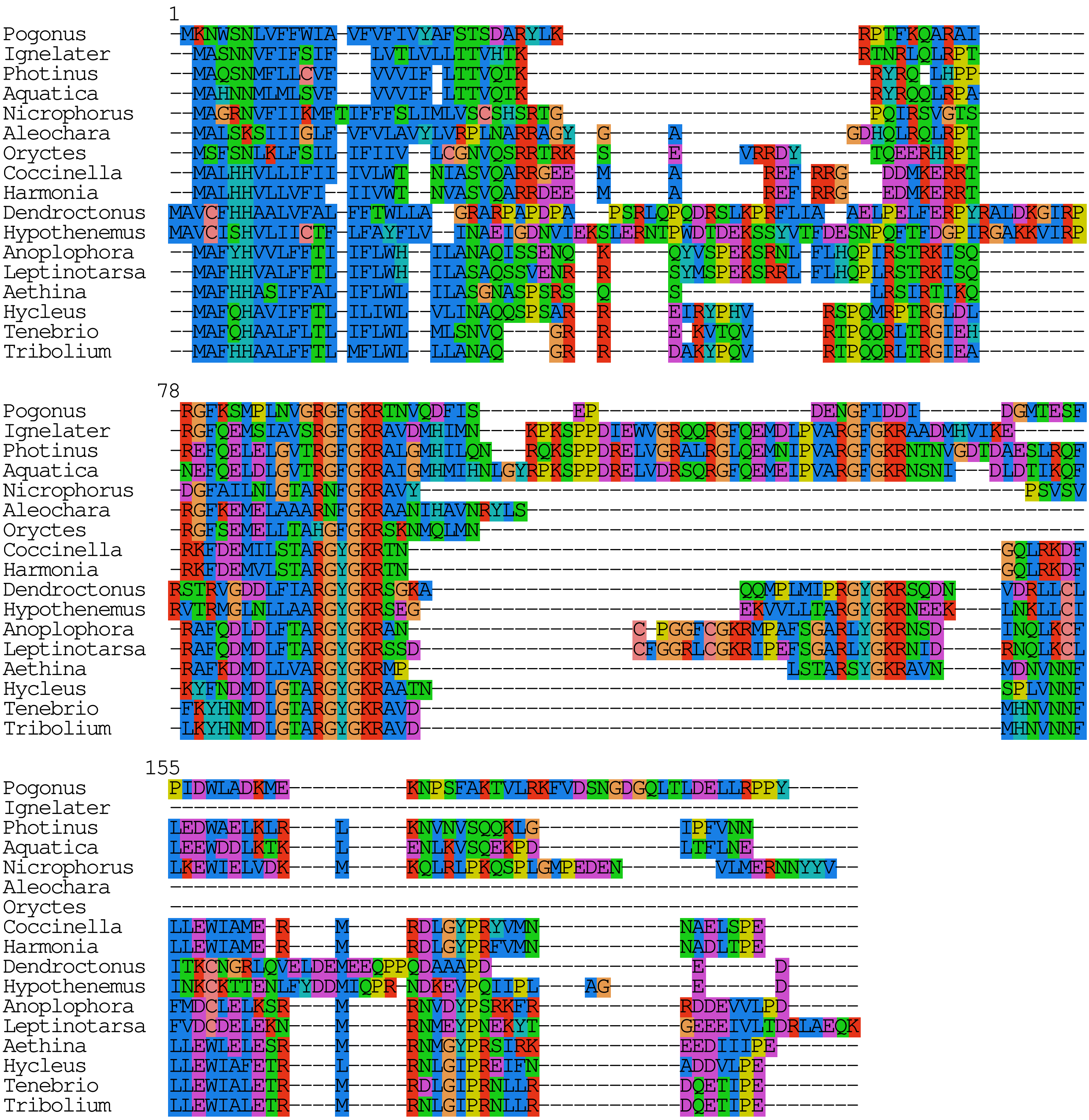


Figure S22. Alignment of allatotropin precursors.

Pogonus
MKTLIILMVISFAFATPYQYIRNRQYQIKHFKNSNQIARTHELPNSLQTLFKSFLLKRCSNLGNDDCSNGYLPE
IGNDDDYLGSGNTPGKRCSNLGNDDC

Ignelater
MNGIILLFGIVLSLDYTWGSPTKRCANMFDEGCINGGVGGAGRDEEWIKDPGNTPGKRCANMFDEGCINGGIGG
SGRDEEWIKDPGNTPGKRCANMFDEGCINEGIGGAGSDEEWIKNPGNTPGKRNLLFKNIPRLYSRSVHKNHQGS
TRRFG

Photinus, pseudogene: has no signal peptide and is not expressed
QIRPKCANLFDESCLNKNLIDSALDEQWLSNGHVPGKRNLNDEGKYTFL

Aquatica
MVKYLVSFLIISISLGSKYVNSARIKRCANLFGEDCINGGTIGGGPDQLWITGNGKQKFLHNLPKVNPQKCVNA
FDTNCINNRRLQEQSNQAWSTYPESTYRKENIQDKKVLNMLYLILKGHSIPTKRCANLMDESCINGGIVGSGDD
EEWLTGDGSPGRR

Nicrophorus
MIRFMVLFILVLGTIDAKPKPCHHRDQACLENQVAIHRPPGIFKSLSDFFNRVDIVTSKRASCSNLYDEGCING
GLPGAGDDEDWLNGNGLGKRASCSNLYDEGCINGGLPGAGDDEDWLNGNGLGKRAGHKDWMNKSGIGKRASCSN
LYDEGCINGGLPGAGDDEDWLNGNGLGKRASCSNIYDEGCINGGLPGAGNDEDWLNGNGLGKRASCSNIYDEGC
INGGLPGAGNDEDWLNGNGLGKRALCSNIYDEGCINGGLPGAGNDEDWLNGNGLGKRASCSNLYDEGCINGGLP
GASEDQAWLTGNGLGKRVNIPQPGYNKNNFKGNKNSKTPIMY

Aleochara
MIRIITLPVVFSAVFLISDGAPCANLFDEGCTYRLLKESAMDQGWLRKNHLGKRTFQEHTQIGIDDFFHRLHNI
LNDRCNNFFDINCIEPNIENHQRVDVNPDAIRAFALQLHEDILNRCAQNAMDKNCNFIQNFTPKDDDDKWIA

Oryctes
MKLLTLILLSVGTINGGYIGKKYCPGLFGDSCASGGLDGSADDEDWLNKPNNPGKRCIANVFDGSCSYGATLPG
AAADEYWKQHGSPGKRCSNVFDESCSNGAVVNGGSDEDFLNGNGNPGKRCSNLFDESCSNAGLIGAGDDESWLN
GGGSPGKRCSNLFDESCSNGGLIGAGDDENWLNGGGSPGKRYLPKQWYKAWGPGGVKTAKRHVSDKVHESLLRK
MAKIRHM

Coccinella
MRKQQFFLMLVLTIHLSQLIECRSYSAAILEEVLRRNSHQNLSPEDVHLFNELYNRLAMPVRKRCANMFDEGCS
NIGIHGSGSDEDWIKVNSPGKR

Harmonia
MKFLFALIMTTIYLFLLVESRSFSSLSFEEAVGRNSHPKLSREDIELFNELYRRLSLPVRKRCANMFDEGCSNI
GIHGSGSDEDWLKDNSPGKR

Dendroctonus
MNRIITLLAISVALSAAMSVPNVYHKTSNSLKQEQLAALINQLLREKQVYQEVKRECLRMGPLGCLNDPRKHLL
TMYNPGKRCANFGDDGCLSGGVQGAGSDSDWLASGFTPGKRCANFGDEGCVTGGVIGAASDSDWIGGGFSPGKR
CANLGDEGCANGGVPGAAADTDWIDGGYSPGKRCANLGDEGCVTGGVSGAGSDSDWISGGFSPGKRCANLGDEG
CANAGVGGAGSDADWLSGDFNPGRR

Hypothenemus
MKCIFIVLAVSVCLSFVRSFPNNPDTYRQSTQLKQKKLSEIIEDLMRDKERYRGSIREYDGCGSAGVPGAGSDS
DWIGGGFTPGKRCANFGDDGCANGGIPGANADQDWLDGGFSPGKRCANLGDEGCANGGVEGAGSDADWLNGNFN
PGRKR

Anoplophora-1
MNFLLAITAVLLGTSQGMYLQRNPHHQQAVARQSRNEPTVLKSLGDFFHRMHAAATKRCAYLLDESCNNGGIPG
AGSDNDWLNQGFNPGKRGLNLFEEGAAYNGLSGSGADSDWLNGGFNPGKRCVNTMDESCSNGGIPGSGSDSDWL

DGGFNPGKRSLNLFEEGIVNKGVSGAAADNDWLNGGFNPGKRCANTMDESCGNGGIPGSGEDRDWLDDGSANPG
KRRLNGIEGVPGSGNDNDWLHGDNTPGKR

***Anoplophora*-2**

MKCLLAITVVLLGSSQGFYVERNPHQQLAIARQSRNQPTIFQSLGDLFHRMHATATKRCAYLLDEACNNGGIPG
AGSDSDWLTQGFNPGKRSSKVLEEEPDNDLNPEPDDDRPNGDFNLGKHCVNAMDESCDNEEIPGAGPERDWLHR
GFHPGKPGLHPFEEDNDVPVAAADGGWLSSFSIVCPNFMDVCGNVGGKRVPGSGADRDWLEEGGGNPGKRRLNG
IPGSGKDNDWLHGDNTPGKR

Leptinotarsa

MNTSTLFLIVLLGFTTAVPLQQWHANKYSSEYHKPNQRGLFSSLGKLFNPSSNKRILNSRGGFSSGERSLNLFD
DSVANSKISGSGSDSDWINGGFSPGKRCANLMGESCNNGGVPGSGSDDDWIHGGFSPGKRSLNLFDDGAANSKI
SGSGSDTEWIDGGFSPGKRCANLMDESCSNGGVPGSGSDDDWIHGGATPGKRSLNLFDDGAANSKISGSGSDSE
WIHGGFSPGKRSLNLLNYASNSKIPGSGSDSDWLNLDGFNSGRRGIKSSEKIGHVFGG

Aethina

MRGIIVVLAMVLAVTLARYVRRHPADVAMTRAKIHLKRNINDPELFKQLGDFFHRMHSASIRCSPGLLDEGCSN
GGIPGSGTDADWINNYGPGKRCVNILSESCDNGGIPGAGSDDDWIHDNSPGKRCANILDEGCNNGGVPGSGSDA
DWINNNSPGK

***Hycleus*-1**

MKAIFIIVLVFLSLANCYYLAPHANRPVKVVPLRNLNDLLYRLNGLALRRCANLFDESCANGGMSGAGADDDWL
NGNTNPGKKRCANLFDESCSNGDINGAGSDDDWLNGDTNPGRR

***Hycleus*-2**

MKFVKILLILLLTSTYAIDIHQSNRKSLKLLKKLINSENYGFLGRLKRRHCANLYDESCINVHLPLAKIDEDWL
RKGFNPGK

***Tribolium*-1**

MKTAFLLLFVLMPAVLCYYVHPNLHYFPARVNYRSAPAKSWGALFHRLQLASKRCANTFDESCINDVINGAGSD
EAFLNGGDNPGKRCVNTFDESCSNGDINGAGSDDDWLHGDDTPGRR

***Tribolium*-2**

MRPVLVLVMIFSVSYARYLEPYNYGYPRPLVDFLNRLSLSDKIKRCGNTFDESCANLPIIGASSDESWLAHSSP
GKRCANVWGESCINGGIIGGGSDQSWLQGDDNPGRR

***Tenebrio*-1**

MKAAFVLLAIALPAAYCFYLQPNYHVPARLGTRNAPVYKSLGDLFHRLHLASKRCVNTVDESCINGGGNDAGND
EDFLNGGDTPGKRCANLYDESCSNGGINGAGADDDWLHGGNNPGRR

***Tenebrio*-2**

MKLSLLIFAVALSTACGLYMYPKHRPRDEEVFQSLASFFNQLGRRANMKRCANTFDESCLNGPIGGATSDENWL
SNGSPGKRCSNIFGSSCVDGGTAGAGADEDFLGGGGGPGRR

Figure S23. Sequences of the various Calcitonin B genes from Coleoptera. Mature peptide sequences are in light blue the conserved C-terminal proline in dark blue, Gly residues that expected to be transformed into C-terminal amide in green and convertase cleavage sites and dibasic amino acid residues anticipated to be removed by carboxypeptidases in red. Signal peptides are highlighted in yellow.

```
              1
Pogonus-1     --MQ--KLNNG-SVIMRTTALILGVFFLCVATLDAAPTPRLIESFKDSVTSIDKCENYLNTKLGQKIKLPVDTIQET
Pogonus-2     --MQ--KLNNG-SVIMRTTALILGVFFLCVATLDAAPTPRLI-----------------------------------
Ignelater     --MKLPTIG-SNG-IMWLVILVAGTLLLISSS---------------------------------------------
Photinus      --MKLPSIT-NNG-IMWFAVILAASSILLSST---------------------------------------------
Aquatica      --MKVPTSA-NNG-IMWLAVAVATSVLLVSST---------------------------------------------
Nicrophorus   --MKSPAFA-FRS-SFVLFLVLAGMLIFLTTS---------------------------------------------
Aleochara     MRFVMQTQTRANG-LS LVVILLGVFLLAATQVSHAA----------------------------------------
Oryctes       --MK-SIMNGSNT-IAVAVLFMAGVLIFMATT---------------------------------------------
Coccinella    --------MVTHS-VAIMLLIVPGLLLRIQNA---------------------------------------------
Harmonia      --------MVLNS-MSFLVFIIPGLLLRIDNV---------------------------------------------
Dendroctonus  MTMKYSSSSGNYG-SL IMLALAALLVLELTT---------------------------------------------
Hypothenemus  --MNTSSPNNRSSCKSIILMALAILLVYELASSVAEAYPARVVEQ--------------------------PNLLFY
Anoplophora   --MKPSMVNSS T-TSVLVLVLVGIVLFVTTT---------------------------------------------
Leptinotarsa  --MRSSMIH-SNG-TTIFMILLVGIVLFEITT---------------------------------------------
Aethina       -----------MQ-RPIFVLLVIGIVLLQVTT---------------------------------------------
Hycleus       --MKPSMNNNNNG-RAFFIFFIIGMVLFEATT---------------------------------------------
Tenebrio      --MKPSMIH-SNG-VSLLVLLAAGIILFEATT---------------------------------------------
Tribolium     ------MVP-NNG-ISLLMLLVAGMILFQATT---------------------------------------------

             78
Pogonus-1     LYMYNSYEQCSIVYALLKIMSPSYYSDSLTNDESQR-----LGILEMLAQLGQNMIR---DDLENSKRGLGFGMDRG
Pogonus-2     -------------------MSPSYYSDSLTNDESQR-----LGILEMLAQLGQNMIR---DDLENSKRGLGFGMDRG
Ignelater     -----------SEAAP---PLPSYYSPLSLEGQNPEYI---LQTLARLRQAIIAE-----EDLENSKRGLDLGLGRG
Photinus      -----------VEAAP-SAPSPGYYAPISVEGQNPEYI---LQALVRLRQALNAE-----EDLENTKRGIDLGLGRG
Aquatica      -----------VEAAP-SAPSSGYYPPLSFEGQNPEYI---LQALVRLRQALNAE-----EDLENTKRGIDLGLGRG
Nicrophorus   ----------TCYAAPHNSRYMGYYGSN QDGQNPEYL---LQTLARIRQAIIAE-----EDLENSKRGLDLGLGRG
Aleochara     ----------PHYPNTLQEYYQNYPRSLDMPNANPEYLFNSIEKLAQLRQQLM N-----ADLENTKRGIDLGLGRG
Oryctes       ----------TSAAPH IFSNPGYYSSLSYENQNPEYL---LQTLARLRQALLAE-----EDLENSKRGLDLGLGRG
Coccinella    ---------------HAASLLPRSPEEDLISRDDPEYI---IRVFRQFRTYYNNIPDDEDDSWTDSKRGLDLGLGRG
Harmonia      ---------------NSASILNSLPEEKIFARENPEYL---LRMYPRLRSTYNIVPDEDDDSWTDSKRGLDLGLGRG
Dendroctonus  ----------TFA SAYPARYSAYYASIPLEE----------QLIERIKQAFIED-----NDLENSKRGIDLGLGRG
Hypothenemus  KTHFHSNCFPIEVNNECVFSYPAYYSPMGLEEQNPNYVN-LQQAIERIRQAFMQN-----NEIENTKRGIDFGLGRG
Anoplophora   ----------NAAPHSPKFYYPAYYSPLSFDGQNPEYL---LQAIARLREALIND-----NDLENTKRGLDLGLGRG
Leptinotarsa  ----------TSAAPR QASYPGYYS---LEAQNPDYL---LQTIARLRQALIND-----DDLENSKRGLDLGLGRG
Aethina       ----------TYAAPHPARYYPGYYSPMQMEGANSEYL---LQTIARLRQALNA------DDLENSKRGLDLGLGRG
Hycleus       ----------TTFAAPHSSSYPGYYSPLSMEGQNPEYL---LQTIARLRQALIAD-----DDLENSKRGLDLGLGRG
Tenebrio      ----------TYAAPHSSRYYPGYYSPLSMEGQNPEYL---LQTIARLRQALIAD-----DDLENSKRGLDLGLGRG
Tribolium     ----------TYAAPHSPRYYPGYYSPISMEGQNPEYL---LQTIARLRQALISD-----DDLENSKRGLDLGLGRG

            155
Pogonus-1     YSAQRAAQHLMGLHAATWAGGPGRKRSGEEVKRMRIGMGMDRGYSGSQAAQYLIGMDAANRIGKRMDSNASNGEPEE
Pogonus-2     YSAQRAAQHLMGLHAATWAGGPGRKRSGEEVKRMRIGMGMDRGYSGSQAAQYLIGMDAANRIGKRMDSNASNGEPEE
Ignelater     FSGSQAAKHLMGLAAANFAGGPGRRRRSEEEA---------------------------------------------
Photinus      YSGSQAAKHLMGLAAANFAGGPGRRRRSVDDA---------------------------------------------
Aquatica      YSGSQAAKHLMGLAAANFAGGPGRRRRSVVDA---------------------------------------------
Nicrophorus   FSGSQAAKHLMGLAAANFAGGPGRRRRSDEDA---------------------------------------------
Aleochara     FSGSQAAKHLMGLAAANFAGGPGRRRRSVSVQEQYANMGDELGV---------------------------------
Oryctes       YSGSQAAKHLMGLAAANFAGGPGRRRRSDENA---------------------------------------------
Coccinella    YSGSLAAKQRMGLASANFAGGPGRRRRNGLEV*--------------------------------------------
Harmonia      YSGSLAAKQRIGWAAANFAGGPGRRRRNGYEA---------------------------------------------
Dendroctonus  FSGSQAAKHLMGLAAANFAGGPGRRRRSDGEA---------------------------------------------
Hypothenemus  FSGSQAAKHLMGLAAANFAGGPGRRRRSDDSA---------------------------------------------
Anoplophora   FSGSQAAKHLMGLAAANFAGGPGRRRRSDDEDA--------------------------------------------
Leptinotarsa  FSGSQAAKHLMGLAAANFAGGPGRRRRSDDDEA--------------------------------------------
Aethina       FSGSQAAKHLMGLAAANFAGGPGRKRRSDDEE---------------------------------------------
Hycleus       FSGSQAAKHLMGLAAANFAGGPGRRRRSDEDA---------------------------------------------
Tenebrio      FSGSQAAKHLMGLAAANFAGGPGRRRRSEEEA---------------------------------------------
Tribolium     FSGSQAAKHLMGLAAANFAGGPGRRRRSEEEA---------------------------------------------
```

Figure S24. Alignment of DH31 precursors.

```
                    1
Pogonus-3/4a        ---------------KRGLGFG-MDRGYSAQRAAQHLMGLHAATWAGGPGRKR--------
Ignelater-2         ---------------KRGLDLG-LGRGFSGSQAAKHLMGLAAANFAGGPGRRRR-------
Photinus            ---------------KRGIDLG-LGRGYSGSQAAKHLMGLAAANFAGGPGRRRR-------
Aquatica            ---------------KRGIDLG-LGRGYSGSQAAKHLMGLAAANFAGGPGRRRR-------
Nicrophorus         ---------------KRGLDLG-LGRGFSGSQAAKHLMGLAAANFAGGPGRRRR-------
Aleochara           ---------------KRGIDLG-LGRGFSGSQAAKHLMGLAAANFAGGPGRRRR-------
Oryctes             ---------------KRGLDLG-LGRGYSGSQAAKHLMGLAAANFAGGPGRRRR-------
Coccinella          ---------------KRGLDLG-LGRGYSGSLAAKQRMGLASANFAGGPGRRRR-------
Harmonia            ---------------KRGLDLG-LGRGYSGSLAAKQRIGWAAANFAGGPGRRRR-------
Dendroctonus-3      ---------------KRGIDLG-LGRGFSGSQAAKHLMGLAAANFAGGPGRRRR-------
Hypothenemus-3      ---------------KRGIDFG-LGRGFSGSQAAKHLMGLAAANFAGGPGRRRR-------
Anoplophora-2       ---------------KRGLDLG-LGRGFSGSQAAKHLMGLAAANFAGGPGRRRR-------
Leptinotarsa-2      ---------------KRGLDLG-LGRGFSGSQAAKHLMGLAAANFAGGPGRRRR-------
Aethina-3           ---------------KRGLDLG-LGRGFSGSQAAKHLMGLAAANFAGGPGRKRR-------
Hycleus-3           ---------------KRGLDLG-LGRGFSGSQAAKHLMGLAAANFAGGPGRRRR-------
Tenebrio-3          ---------------KRGLDLG-LGRGFSGSQAAKHLMGLAAANFAGGPGRRRR-------
Tribolium-3         ---------------KRGLDLG-LGRGFSGSQAAKHLMGLAAANFAGGPGRRRR-------
Ignelater-1         ------------KRNIKSVDFG-MGRGHSGTLEAQYRMALESAFSPNGPGRKKR-------
Hycleus-2           -------------KRYLQLSFG-LDRGSSGNFENFTRCLQLIASSPYGPGRRKR-------
Tenebrio-2          ---------------KRFVSFG-VDRGSSGKVENLTRCLAFRASSPCGPGRKKR-------
Tribolium-2         ---------------KRFLSFG-VDRGSSGKVENLTRCLAFRASSPCGPGRRKR-------
Pogonus-1/2         ------------RLRRTIPDFG-LSRGYSGNLEAAHRLGYDRAMDPQGPGKR---------
Aethina-2           --------------KRISLDLG-TDSSHPAKLQNDILNMNELLRNYG--------------
Hycleus-1           --------------KRARPDLG-MDRGFSGRLQNSQLVAKYYGDLRG--------------
Tenebrio-1          --------------KRGGPDLG-MDRGYSGKLQNSQLLAKYYGDLRG--------------
Tribolium-1         --------------KRVRPDLG-MDRGYSGKLQNSQLLAQYYGQLRG--------------
Aethina-1           ---------------KRGIDFG-INRGFPPLWKKCMLTKMNALKTYG--------------
Leptinotarsa-1      ---------------KRVIDLGTQSRGYSGKMENMYLTNADMLRRIGKR------------
Pogonus-3/4b        --------------KRMRIGMG-MDRGYSGSQAAQYLIGMDAANRIGKR------------
Dendroctonus-2b     RVRKQSLVNCAEIMSKAQLDFG-TRQSDAGSIENCLMTLRTELNHLGK-------------
Hypothenemus-2      ---RRPRINCSQFFSKGGLDFG-TRQNDPGSIENCLRTIRTELHHFGK-------------
Anoplophora-1       ---------------KRKTDFG-TDRGYSGQQEHNMRMNKYLLEYVG--------------
Hypothenemus-1      --------------LKRQADFG-SQRGHTGRQEYALRYWSHLG------------------
Dendroctonus-1      ---------------KRTADFG-SQRDHTGQREYALRYWTHLG------------------
Dendroctonus-2a     ---------------KRTADFG-SQRDHTGQREYALRTITEKPELVPESNTMEPRVRKQSL
```

Figure S25. Alignment of DH31 and other putative neuropeptides encoded by transcripts from Coleoptera DH31 genes. Numbers behind the names indicates the transcript number (some species seem to have only one transcript). When this number is followed by a letter it indicates the putative peptide on that particular transcript. For example Pogonus-3/4a indicates DH31 itself that is coded by two transcripts in Pogonus and a refers to the first putative neuropeptide on this transcript. Pogonus-3/4b refers to the second putative neuropeptide on this transcript. Note that sequence conservation is very strong for DH31, but much less for the other putative neuropeptides encoded by these genes.

```
             1
Pogonus      -----MRVPVMYLVCAATLLVSS-RAAPVDTDSNDPETSMG--IGSAASINGQDPSETL-AYLLPRLSTRYRPTEQW
Ignelater    -----MRVPV-YLVCVALILCVNCE---------EQTNYY---GRRIQPVDLTADQETV-TYLLPKLTAKYRPNNEW
Photinus     -----MRVPV-YFVCAALILYVKSE---------DRTN Y---DRRIEPLDLTADQETV-SYLLPKLTAKYRPNTEW
Aquatica     -----MRVPV-YFVCAALIFYVKGE---------DLSN Y---DRRIQPLDLTADQETV-SYLLPKLTAKYRPNTEW
Nicrophorus  -----MNVPV-YIVCAALIVAVNSE---------ENP LF---GRENEPMD----REAM-GYILPKLMPRYRAISNK
Aleochara    -----MRVPV-YIVCAALIVAVNSE---------ENY YP---GRQQE-------ADAL-GYILPRLMAKYRPMRPW
Oryctes      -----MRMPV-YLVCAALAVAVKSE---------ELPSLY---GRQLEPLEVNSEQDAL-RYIFPRLTSRFRASKNW
Coccinella   -----MRWSI-YLACAALVISIKAD--------ENSVDYY---GSFLEPINIAAEQETLNNYLLPKVQPKYKP--EW
Harmonia     -----MHWPV-YLAFAVLVIATKAE--------DNSADYY---GSFLEPINIAPEQETLNNYLLPKIQPKYKP--DW
Dendroctonus -----MRAPI-YLVCAALVVAVRSE---------PNENYM-----GLESVNVAADPETI-NYLLPKLAAKYRLNSDW
Hypothenemus -----MRAPL-YLVCAVLAWTAHGD------DQGQQQNYM-----DLEPVDVAADPETV-NYLLPKLTAKYRPNNEW
Anoplophora  MGAHIMRMPV-YLVCAAALVA  VK------GE EGTNY---FGRQLEPVDVAADQETI-SYLLPKLAAKYRPSNDW
Leptinotarsa -----MRAPL-FVLCAALVVAIRSE------DTSAVNTNF--FNRQLEPINMAADQETI-SYLLPKLAAKYRPNSDW
Aethina      -----MRASF-NLLCAATLMFALASCAELNNEATGTGNYYGDAGRSLEPLNVAADPETI-NYLLPRLTAKYRPNNDW
Hycleus      -----MRVPL-YYLLWAILIVTVKT------NG ETSNYY--GGRFLEPIDVAADQETV-SYLLPKLTAKYRPNSDW
Tenebrio     -----MRVPV-YLVCAALVVA  VK------SE DH---N--YGRLLEPIDVAADQETV-SYLLPKLTAKYRPNNEW
Tribolium    -----MRVPV-YLVCAALVVV  VR------SE ERTNYY--GGRYLEPVDVAADQETV-SYLLPKLAAKYRPNSEW

             78
Pogonus      NTMAEQEFYLDNNGF--------PEQE-----------PE---ERFKRA------GNGPSLSIVNPIDVLRQRLLLE
Ignelater    NGVTDPRFYVLTEME-SNDIDY---QEPL--------AGEG---RIKRQGL----NNGPSLSIVNSLDVLRNRLLLE
Photinus     NGVKDPRFYVLTEAE-SNDIDF---QEPL--------IGEG---RIKRQGL----NGGPSLSIVNSLDVLRNRLLLE
Aquatica     NGVKDPRFYLLTEME-KNDIDY---QEPL--------IG-----RIKRQGL----NNGPSLSIVNSLDVLRNRLLLE
Nicrophorus  QWMAPYY---------ADSNNNNELD---F----------GQEARNKRA----GLIDGPSLSISNSLDVLRNRLLLE
Aleochara    AYYADLD---------ANTIDTIPQQLPQFQRTLESQSQEDDRDKRKRAGLSLREVNGPSLSISNSLDVLRNRLLLE
Oryctes      DDSRYYD--IPTDYQ-LNTIDEIPFSQ----------EGRIKRANFNDKPSSVKQFRGPSLSIVNSLDVLRNRLLLE
Coccinella   RRLGGTGLYLVTDDL-NTDSER---LN-----------PET---RIKRAEF---MRPRGSLSIVNSLDALRNKLVME
Harmonia     RRLGDPSVYLMSDDL-NSDSER---LRDV--------PSET---RIKRAEF---LRPRGSLSIVNSLDALRNKLVME
Dendroctonus RGVSDPKFYVLTEME-SNDIEN---QEPT--------AEEDLNLRTKRQSRIG-RNAGGSLSIVNSLDVLRNRLLLE
Hypothenemus RTVTDPRFYVLTEMIGSNDIDN---QIGY--------FDENA--REKRQS SRIGDAGGSLSIVNSLDVLRNRLLLE
Anoplophora  SGVTDPRFYVLTEME-SNDIDN---QNDL--------GLDS---RIKRTGR--LGEQGGSLSIVNSLDVLRNRLLLE
Leptinotarsa AGVTDPRFYLLTEME-SNDFDN---QETS--------NLET---RTKRAGR--IRESGGSLSIVNSLDVLRNRLLLE
Aethina      TGVTDPRFYLLTEME-SNDIDN---QNNL--------SPEA---RSKRAGR--LGEPGASLSIVNSLDVLRNRLLLE
Hycleus      SGVTDPRFYVLTEME-SNDIDH---QNGL--------RS-------KRAGGIGGGEAGASLSIVNSLDVLRNRLLLE
Tenebrio     SSVTDPRFYVLTEME-SNDIDN---QNKL--------GPEL---RSKRAGA--LGESGASLSIVNSLDVLRNRLLLE
Tribolium    SGVTDPRFYVLTEME-SQDIEN---QNKL--------SPES---RSKRAGA--LGESGASLSIVNSLDVLRNRLLLE

            155
Pogonus      IARRQMRESQKQVAENRKLMKTIGKRGRFLRQPHDMRRPNPLGFDNM---
Ignelater    IARKKQLEG---MNRNRNLLSSVGKRATFGHRGALG-YDNNL--------
Photinus     IARKKQLEG---MNRNRNLLSSVGKRAILGHRGLVP-TFDNF--------
Aquatica     IARKKQLEG---MNRNRNLLSSVGKRAVFGHKDVIA-GYDNF--------
Nicrophorus  ISRKKALLG---ANYNRNMLCRIGKRFEN NKRRCE-DSEKYVEDRFMV-
Aleochara    MSRKKELEG---ASRNRNILCKIGKRAFD-GKRRCE-EPEKYVDDKLGEA
Oryctes      ISRKKALEG---ANRNRNYLCKIGKRSPLFDQKQCE-DDNIKAFL---P-
Coccinella   MARKKTQQN---AERNRQFLQSFGKRSYAVHR NID-RFNNM--------
Harmonia     IARKKTKQN---AERNRQFLQSFGKRSYPVHRRSMD-RFNNM--------
Dendroctonus IARKKAKEG---ANRNRQLLMNFGKRAFIQRIPRAY-FQRQI--------
Hypothenemus IARKKAKEG---ANRNRQLLMNFGKRAYLPTRRLFV-NRKI---------
Anoplophora  IARKKAKEG---ANRNRQMLMNLGKRAFLQR PAGV-YDNKM--------
Leptinotarsa IARKKAKEG---ATRNRQMLMSLGKRAYFSRRLYGD-DE-----------
Aethina      IARKKAKEG---ASRNRQILMHLGKRAFYHR NTGI-FDNKV--------
Hycleus      IARKKAKEG---ATRNRQILLSLGKRAFLQSRNNGR-YDNNV--------
Tenebrio     IARKKAKEG---ANRNRQILLSLGKRAFLQSRASGT-YDNNV--------
Tribolium    IARKKAKEG---ANRNRQILLSLGKRAFLQSRASGT-YDNNV--------
```

Figure S26. Alignment of DH47 precursors.

```
              1
Coccinella    -----MRWSIYLACAALVISIKADEN---------SVDY--YGSFLEPINIAAEQETLNNYLLP
Harmonia      -----MHWPVYLAFAVLVIATKAEDN---------SADY--YGSFLEPINIAPEQETLNNYLLP
Dendroctonus  -----MRAPIYLVCAALVVAV--RS-------EP-NENY----MGLESVNVAADPETI-NYLLP
Hypothenemus  -----MRAPLYLVCAVLAWTA--HGD----DQGQ-QQNY----MDLEPVDVAADPETV-NYLLP
Anoplophora   MGAHIMRMPVYLVCAAALVAV--KGE----E----GTNY--FGRQLEPVDVAADQETI-SYLLP
Leptinotarsa  -----MRAPLFVLCAALVVAI--RSE----DTSAVNTNF--FNRQLEPINMAADQETI-SYLLP
Aethina-1     -----MRASFNLLCAATLMFALASCAELNNEATGTGNYYGDAGRSLEPLNVAADPETI-NYLLP
Aethina-2     -----MRASFNLLCAATLMFALASCAELNNEATGTGNYYGDAGRSLEPLNVAADPETI-NYLLP
Hycleus       -----MRVPLYYLLWAILIVTVKTNG----ET---SNYY--GGRFLEPIDVAADQETV-SYLLP
Tenebrio      -----MRVPVYLVCAALV-VA-VKSE----DH------N--YGRLLEPIDVAADQETV-SYLLP
Tribolium     -----MRVPVYLVCAALV-VV-VRSE----ER---TNYY--GGRYLEPVDVAADQETV-SYLLP

             65
Coccinella    KVQPKYKP--EWRRLGGTGLYLVTDDLNTDSERL-APYRPNLRKRYYESYIDD-------RNVR
Harmonia      KIQPKYKP--DWRRLGDPSVYLMSDDLNSDSERL-EPYRSNIHKRYYESYVDD-------RNVR
Dendroctonus  KLAAKYRLNSDWRGVSDPKFYVLTEMESNDIENQTPEQPRRIRNKRYNNY-------------S
Hypothenemus  KLTAKYRPNNEWRTVTDPRFYVLTEMIGSNDIDNQGRKPKRIRSKRY-NY-------------T
Anoplophora   KLAAKYRPSNDWSGVTDPRFYVLTEMESNDIDNQ-ITKPKKIRNKRYNYDLN------------R
Leptinotarsa  KLAAKYRPNSDWAGVTDPRFYLLTEMESNDFDNQ-VANNRRIRNKRYDFDVYEPRQVRYDFGEE
Aethina-1     RLTAKYRPNNDWTGVTDPRFYLLTEMESNDIDNQV-SHIRK-RAD-----------------DR
Aethina-2     RLTAKYRPNNDWTGVTDPRFYLLTEMESNDIDNQN-HMGGKLKKR-----------------QN
Hycleus       KLTAKYRPNSDWSGVTDPRFYVLTEMESNDIDHQ-V--GRNVQRR------------------S
Tenebrio      KLTAKYRPNNEWSSVTDPRFYVLTEMESNDIDNQ-MPSERSIQRR------------------S
Tribolium     KLAAKYRPNSEWSGVTDPRFYVLTEMESQDIENQ-VPSERSIQRR------------------S

            129
Coccinella    PSLSIKAPIDALRKAMLHKNWERNVQKNRELLQRL-----
Harmonia      PSLSIKAPIDALRKAMMYKNWEKNVQRNRELLQRL-----
Dendroctonus  PTLSVQAPIQALRNLYDRSNYQQRISQNRRLINQLNSS--
Hypothenemus  PTLSVQAPIQALRNLYDRSTYNQRAFQNRQLIRQLTSS--
Anoplophora   PSLSIQAPIQSLRNAYAKEKLRQRLEDNRRFLANLVDIQ-
Leptinotarsa  PSLSINANIQNLRQAYAKTKLRERLAANRKFLENLRDSQ-
Aethina-1     AVLSIQAPVRALYDSMQKNKMRKLLLANRQLLNNLRDQYN
Aethina-2     HVLSFQEPRHMLNRHLEKQKHERQSELNRNFLNNLVDYY-
Hycleus       PTISIAAPIDVLRKTWQQEHARKQMERNREFLSRLT----
Tenebrio      PTISITAPIDVLRKTWEQERARKQMVKNREFLNSLN----
Tribolium     PTISITAPIDVLRKTWAKENMRKQMQINREYLKNLQ----
```

Figure S27. Alignment of DH37 precursors.

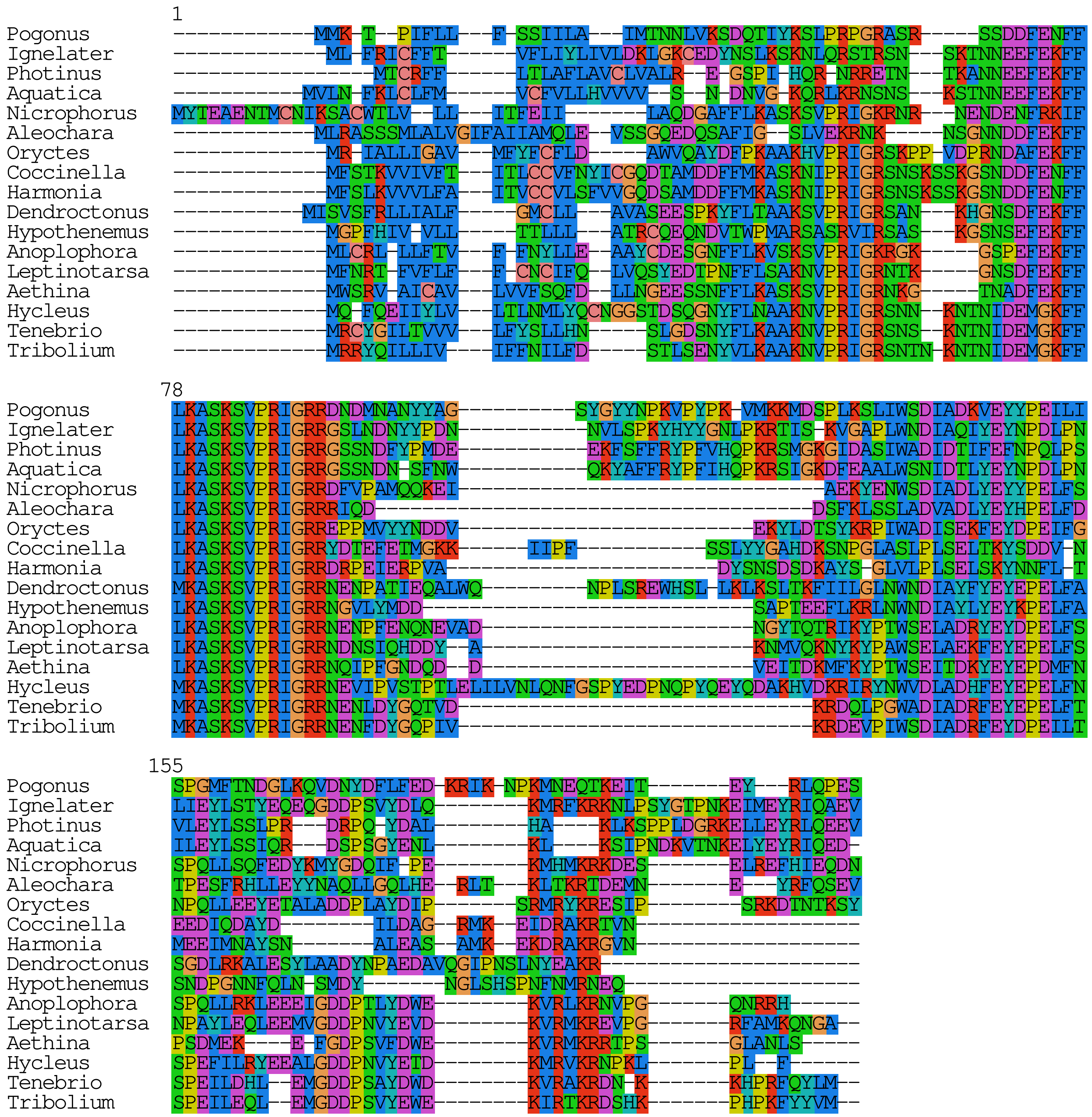


Figure S28. Alignment of ETH precursors.

```
             1
Pogonus      --MCVWRVVR---FTIYLLVIMVSLASAREFVDKRTPEDRQSWFMGSRYGRSSNPGGGGGG---
Ignelater    --MNTGKVLV---LAAYFFTALLAVAVARDYYSTDKRAQDRPFMMSSRYGRAGGGTNAANG---
Photinus     --MNAKTIFV---LAAYFFTALVTVGAVRDYYLSEKKAQDRPFLMSSRYGRGGGGTSGTNG---
Aquatica     --MNAKKIIV---LAAYFLTALLTVAETRDYYATEKKAQDRPFMMSSRYGRAGGSTTSSNG---
Nicrophorus  -MMNAQKIII---LAAYFLTALIAVTATKGYYHNDKRANDRPFMMGMRYGRADKA--AKTA---
Aleochara    MVAHQNGCKF---LITYICNPLIFLCL---------FCAGKAFNPGPRFGRSYMSGGGGGSKKI
Oryctes      --MHVSKIVL---LAAYFLTAVIAITANKETYTQQKK-LGKDFLLGMRYGRSRDGTGGSTK---
Coccinella   --MFSNGLSVQMNVILLIIGILISFLSY------------NEAILTTRYGKKNINSGEI-R---
Harmonia     -----------MNILLSIIGVSIFLLDT------------KEALLTTRYGKKNINTDDI-R---
Dendroctonus --MLVRKVPV---IFVFLVSIFITVVNADMESIKRIPS--HSFKQMMRYGRSSAN---------
Hypothenemus ---------------------MVSFVIADMESNKRIPGAAHAFQQMVRYGRSGNN-----K---
Leptinotarsa --MHIRRIIV---VLLYCITVLLTAVK--AY-SDKR--IPEKFKSMVRYGRSSKSSSNERD---
Aethina      --MYAKKALV---LLLYFLTVFLTFAVAKGYLSDKRAQA-NSFQTMMRYGRAGSTNNYYKD---
Hycleus      --MHPRRVFI---VLIYIFTVLLSTTLAKHYASDKRVQSPSSFKTMMRYGRSGISNANGKE---
Tenebrio     --MHARKVIV---LLVYILTVLVSVAVTKRYTSDKRVQNLSTFKTMMRYGRAGPN-PNEKE---
Tribolium    --MHARKLIV---VLVYILTVLVSVAVSKRYTSEKRVQNLATFKTMMRYGRGGPS-PNNKE---

            65
Pogonus      -TRHLIVTPRRDDRFHPGSRYGKRTVSTSGASGMRSLASVTTNLRSNEENTNDLGSETFATIGD
Ignelater    -GKHVVVAPR-TDKFFLASRYGKRSSML--SS-AP-LAY-------------------PADSLP
Photinus     -AKHVIVAPR-TDKFFLASRYGKRSSTM--SN-AP-LSY-------------------LTDTLP
Aquatica     -PKHVIVAPR-TDKFFLASRYGKRSSVL--SS-PP-LAY-------------------LPDSLP
Nicrophorus  -GKHVIVAPR-NDKFFLASRYGKRSGGEMISNAAQ-AAL-------------------VFPVPP
Aleochara    SGTRTMVAPR-NERFELGSRYGKRGDLAEVP--MP-LIYPI-------------SAGAVDTKQF
Oryctes      -----QKQHR-SDVFYLGSRYGKRSSDS---PGVP------------------------LMYSQ
Coccinella   -PRSINN----VPSFFVGPRYGKRMAWSPGENELS--------------------------SPL
Harmonia     -PRSVKN----IPSFFVGPRYGKRMTWSTREDQLP--------------------------SPH
Dendroctonus -NEKINVNPR-ADLFYLGPRYGKRAGPTLAK-------------------------------LD
Hypothenemus -DNKIKIFPR-ADVFYLGPRYGKRSRELEDN-------------------------------VR
Leptinotarsa -TKPINV-RR-GDAFLVGTRFGKRSSWN---PNAS-LVY--------------------SVSAS
Aethina      -GKYVKVNPR-ADAFHLTTRYGKRSSWS---PNSS-LVY--------------------PVSSP
Hycleus      --NKVTIHPR-ADAFFLGPRYGKRATWN---PNMS-SGY--------------------SDALP
Tenebrio     --SKVNIHPR-ADAFFLGPRYGKRSSWS---PNAS-LVY--------------------PVSTP
Tribolium    --NKVNIRPR-ADAFFLGPRYGKRSGWS---PNAS-LVY--------------------PVSTP

           129
Pogonus      LSGERQ--NVECSYTGVADLYRCHIHVRKENTSGEFLSN---
Ignelater    LCGTDE--DLSCAYTGFSNFYRCDQKKSSISDAISSVN----
Photinus     LCGNVEDLDLSCAYTGFSNLYRCAQKKETPSIDLSSDN----
Aquatica     FCGNDE--DLSCAYTGFSNLYRCAQR----------------
Nicrophorus  ICGSDE--DLKCTYSGISNIYRCIQSYRKDSAQGNGIMTTSI
Aleochara    QCGKGD--EIMCSYTGILNIYRCNQRCVGCILTTL-------
Oryctes      LCGNDN--DIYCDYTGVSNLYRCIQSFRNDGSIEDLITST--
Coccinella   PCTTFE--DLSCDYTGISDYYRCYLRRDQDEE-DNFSDSN--
Harmonia     PCTTYG--DLSCDYTGYSNYYRCSLRRDREGEEEDFADSY--
Dendroctonus --TVYDSTFMPCTYTGVADLYRCDTSRSTMREDRRK------
Hypothenemus -NEQDDDESFPCIYTGVTNLYRCNMSRYH-------------
Leptinotarsa PFQKHE--PLSCTWVGLSNFYKCRSRKDSLTTMD--------
Aethina      -CVEDE--DLSCTYTGISDLYRCTTRKDSTAGNEEI------
Hycleus      LCGFDE--DLSCTYTGIQDLYRCSPR----------------
Tenebrio     LCGLDE--DLSCAYTGISDLYRCTPRKGRESDFSTSSN----
Tribolium    LCGLDE--DLSCAYTGISDLYRCTPRKGESEEFTTSSN----
```

Figure S29. Alignment of RYamide precursors.

```
Pogonus       MGCRNILRIILLTIIIC-VLLSHRQHTVKGTP-T--NIGDNNHQTRNRPN-RFGT-RLTSQYVR
Photinus      MFQARLPTGLFAISSIYLLLFLNHSQNANSFVSA-LNNMEALHR-PHRPLPAASA-RLAIHYKK
Aquatica      MFMVKGRSRVLFILSAL-ILFIIHNQNVNSA--T-AAAIDGSHR-QHRPIPRVSA-TIPLRFNK
Ignelater     MSSIRILTGILIIILSAF-ILLLHQKQHIQGVSAA-TVNMENNHR-PHRPVPRVAA-RLPVHYSK
Nicrophorus   MSQ-TYFMMKLLLVAMC-LILMACNDGASAGPAGASVPTEANRRIRSRPFLRVSP-RTIGQYAR
Aleochara     MICKGIIRS-FLLCFIF-LVVCNHMQRVSAAASANVQPNENSHRLRPRPYLRIPQ-RAFNQYAR
Oryctes       MVVHKIFTVFVIIFFVY-TLKLDNSQFVAGA--PSTNTLESIHRHRSKPLLRLLP---KSQYPK
Coccinella    --MAKVMTAIFFILSVY-LLFIHQLHIVSSAIPSY-----------SNRLEKLSLGKFKNRRIN
Harmonia      --MAKIMTGIFCILSIY-LLLIHQIQIVASTIPSR---------KLFGRFDRLHLGEISNRKVH
Dendroctonus  --MKNAITSYFLIIAICFFLILCQMNHIIAESSN---GNVELP--RNRPSGLLKYPYRLVQYRR
Hypothenemus  --MNHLLTNPFVLISICLLITCHCLTITSNNNIPGGLPR-------TLSTNTVYHQRAHTQYNR
Anoplophora   --MKSFFTGMFLLTSLY-LFLTYQFGFTSAT--TGGPTNQEVHRPRARILGRFA--RPITQYAR
Leptinotarsa  MVLRSLFTGFFIVSSLY-LLLTYQFGLSLGA--PGRGQN--NHRSRNRGFGRHPI---SPQYAQ
Aethina       --MRSLLTGMFLITSLY-LLFIHQLQSV--Q--PAHVPSADLHRVRARPINRFNK-PLLNQLAR
Hycleus       ---MKISTGFFILSSIY-LICINQLTIVSAS--TGSSPNLENRRQRPRPFVRVPLGRPPVQYAR
Tenebrio      MGMKSVFTGIFIISSMY-LLFIHQFHNASAA--PGNVNNLDSHRPRGRPFTRLTP-RTSNQYAR
Tribolium     MGMKSFFTGVFLISSVY-LLFIHQFQNVSAA--PGNANNVDSHRLRARPFARLTP-RT--QYSR

             65
Pogonus       TRVEP--D---FI-LDDDDLFDLTKRQS---DDYGHMRFGKR-E--FDDYGHMRFGRSGTD---
Photinus      VHP-EL-S---DI-LDEEDLFDLSKRLV--TDDYGHLRFGKRGED-FDDYGHLRFGRGGTENK-
Aquatica      IRTETL-S---DL-FDDDDLFDLSKRLV--TDDYGHLRFGKRGED-FDDYGHLRFGRNGRDKEL
Ignelater     IRTESL-S---DF-LDDDDLFDLSKRQV--ADDYGHLRFGKRGED-FDDYGHLRFGRSGGEKK-
Nicrophorus   FKP-DLGD---FI-VDDDDFFELNKRQN--SDDYGHLRFGKR-ED-FDDYGHLRYGRSGDSSRR
Aleochara     LKADE-------F-LDDDDFFEMNKRQT--SDDYGHLRFGKR-DE-FDDYGHMRFGRSGSDTNN
Oryctes       IKA-DFSD---FI-VDDDDLFELSKRQT--SDDYGHLRFGKRGDEQFDDYGHMRFGRSGNSD--
Coccinella    QRSDFLFN---DF-IDEEDLDMTNKRQI---DDYGHMRFGKRGEDQFDDYGHMRFGRSF-----
Harmonia      PKSDFIFN---DF-IDDEDLETNNKRQV---DDYGHMRFGKRGEDQFDDYGHLRFGRSF-----
Dendroctonus  LKPHAIIN---DFDEDDDDVFNLSKRQQTNSDDYGHLRFGKR-EEQFDDYGHMRFGRNGGERK-
Hypothenemus  LKPESFFVPNDLVLLDDDDLFDLSKRLPTASDDYGHFRFGKR-DPVFDDYGHMRFGRGGAD---
Anoplophora   LKPEPYLN---DF-IDDDDLIEFSKRQT--SDDYGHLRFGKRGEESFDDYGHMRFGRSGTDQ--
Leptinotarsa  MKNEPFVS---DF-IDADDFFEISKRQQNNPDDYGHMRFGRKGEEISDDYGHMRFGRSDIENK-
Aethina       IK-EPVDA---FI-VDEDDLFDLAKRQT--SEDYGHLRFGKR-DEQFDDYGHMRFGRGGD----
Hycleus       AKSEPFGD---FI-VDDDDLFELSKRQT--SDDYGHLRFGKRGEDSFDDYGHMRFGRSGSD---
Tenebrio      IKPEPFNE---FI-VDDDDLFELSKRQT--SDDYGHLRFGKRGEEPFDDYGHMRFGRSGGDK--
Tribolium     IKAEPFNE---FI-VDDDDLFELSKRQT--SDDYGHLRFGKRGEEPFDDYGHMRFGRSGSD---

           129
Pogonus       -
Photinus      -
Aquatica      S
Ignelater     -
Nicrophorus   -
Aleochara     -
Oryctes       -
Coccinella    -
Harmonia      -
Dendroctonus  -
Hypothenemus  -
Anoplophora   -
Leptinotarsa  -
Aethina       -
Hycleus       -
Tenebrio      -
Tribolium     -
```

Figure S30. Alignment of Sulfakinin precursors.

```
             1
Pogonus      -----MWIWILLAALINQ-SFISSSAAEPIGSEYQEEAPDDSMYLYLEERSVKDN---------
Ignelater    ML----RLIIVVVLIHISSAIPMFDDS---YD--------DEPFLYMDSRPSKYR---------
Photinus     MH----RLMILILLVQLSSIEVFSEDA---YD--------EDPLTFLESHISKYR---------
Aquatica     MY----RLVVVALLILLSSLEVISEES---YD--------EDPVLFLEPFSS------------
Nicrophorus  ---MSTMLNLLFLLAALEPIWAYASPE-EGYLLPQTSGEDDDLYLYVDTRSESEQ---------
Aleochara    ----MAMITLIIGILLVQ-ALRFASTAEEAFLGYPLQTEEDEGYLYMDSRADPGY---------
Oryctes      MLAHP-ITALLFHFLIMT--TAYSANS---IYQQPSKTKGEQLFMNLRSNYDTERSTRRVFAQN
Coccinella   -----MYISALLVLVLTQYTWAYNDDN---YPFSESYDP---T-LISAEDY-ADQ---------
Harmonia     -----MYISGLLVLVLTQYTWAYNDDN---YPFSDNYDP---N-IIPFEDY-TDQ---------
Dendroctonus ----MGLSVILMALLLLQSALSLTDDSFEGYEIFGPDEE-----------ADME----------
Hypothenemus ----MGLSSVFLLLFLLRFAVSINDDSIEGFELFVPEDE-----------SDLE----------
Leptinotarsa -----MLPIFLFIFFLVQSSFANLEES---YHETPDSYS-----YYPDDLDSDD----------
Anoplophora  -----MIIIPIISILLIQSSLAFLEDS---YPETFDSNTY------PEDLFDND----------
Aethina      MFFF-----PLVILFLVQSTWGYVDET---YYPGENVDNY----LYSEEVTENEN---------
Hycleus      MSPIPLICIILLLLHHHHTVWCYNNDD---LYQQFPPDPYDSI-YYSIE-DYFDE---------
Tenebrio     ---------------------------------------------------QPPP---------
Tribolium    MVPF----AILILTLIIQLASGYNNED---FYSDNF--------------DGFEE---------

            65
Pogonus      -----------T---------------DDIEARRRSAL--------DKNFMRFGRGGNTNDLY-
Ignelater    -----------PS--------------R--------LLQDKDA------FYRVSRAHEDSYA--
Photinus     -----------PY--------------K--------TFHDKDRGFPDFNYLRYSRGRSESIL--
Aquatica     ---------------------------------------------------RYTRSRSESLL--
Nicrophorus  ---------------------------RSSEPSRRSTL--------YKNFARLGRGDLPANY--
Aleochara    -----------SPSML-----------KQGETRRRSTL--------DKNFARFGRSGLGKSKLS
Oryctes      PIRFGGRRSDIPPLVGKAMEYEAMLDNEVGDRERRAFSKGNHMRFGRANFLRFGRSRSNTNN--
Coccinella   ---------------E-----------DIFEPA--EKR--------ANNIPRY-------EH--
Harmonia     ---------------E-----------DIFEPA--EKR--------SESLGRY-------EQ--
Dendroctonus ---------------------------P--YMDRQSRG--------NDKFLRFGKAFWDYES--
Hypothenemus ---------------------------S--YIDRQTRN--------NDKFLRFGKAFWDYSD--
Leptinotarsa ------------------------------NEM--EKP--------NNHFLRLARSDGNFD---
Anoplophora  ---------------------------NDIEVQ---RR--------SNHFLRFGRNNGKYEN--
Aethina      -----------PNPPQ--------------RRN---NN--------NNNFLRFGRSEK--KY--
Hycleus      -----------PDEIY-----------RHFY-N---DK--------NDNNDRIATIIKRHDN--
Tenebrio     -----------PPPAS-----------SVLTPL---LR--------GGYHRRVRG--T--NY--
Tribolium    -----------PSEEV-----------SDMEVR---RR--------NSNFLRFGR--S--GP--

           129
Pogonus      -LQK--HILDPDDVDYNDE---------YNRDQR---TSNKNGNFIRFGRGKAGDFIRFGRNPT
Ignelater    PLI---DEIDTEFEDFNDE---------FARPTRS---QDKNDNFVRFGRGN-SNFVRFGRDSY
Photinus     PFM---DDLESEYDD-EEQ---------FARPTRS---QDKNDNFVRFGRGN-SDFIRFGRDPY
Aquatica     PLM---DDLESEYDD-DDQ---------FARPTRS---QDKNDNFVRFGRGN-SDFIRFGRDPY
Nicrophorus  --------------EMEEG---------YDRPTRV--LGDKSDQFIRFGRGN-SDFLRFGRD--
Aleochara    YLNEDNRLKDMMYMDEDADETLTNNGDSGRFLERNSRSGERNDQFIRFGRGK-SDFVRFGRADS
Oryctes      -------------NSNRND---------YNRLTR----TNNSQFLRFGRGR-SDFLRFGRSEE
Coccinella   ---------R---RSSNDP---------DPRPTRGAKM-LRSDYFVRFGRSK-QDFLRFGRDPN
Harmonia     ---------R---RSSNDP---------DPRPTRGAKM-LRSDYFVRFGRSK-QDFLRFGRDPP
Dendroctonus --GN--E-----APYSDKA---------DQRPPRTG---------------------SLPDNPQ
Hypothenemus --AG--N--DAAAMYIDKE---------APRPSRTGRQ-GNNEHFIRFGRSN-KDFLRLSGNPQ
Leptinotarsa ---------------EYDD---------EDGPIIGKK-NLKNDHFIRFGRSSHDRCLRLGRDLE
Anoplophora  ---------NEVGNDDYED---------FARPTRSGRI-EKNDHFIRFGRGKHQEFLRFGRDPQ
Aethina      ---------DTDYEDYIED---------FARPTRSGRT-DKNDHFIRFGRAG-QDFLRFGRSQV
Hycleus      ---------SLSDLSYDSK---------FPRPTRSGAIGDKNDHFIRFGRSK-QDFLRFGRRST
Tenebrio     ---------DVEYDDFNED---------FARPTRSGKT-EKNDHFIRFGRSK-QDFLRFGRDQH
Tribolium    ---------NYEYEDYGED---------FARPTRSGKI-EKNDHFIRFGRSK-QDFLRFGRNQP
```

```
             193
Pogonus      Y-LRDIRGRDQNMIRFGRSVPLKRSK--------RQVNDM-YSQEEEKR-RTNNMLRFGR---N
Ignelater    KH------KNQDFVRFGRSISESEMP------KKRSRRDV---DESKKRNDRDNFLRFGRGGGD
Photinus     KYGREMRANDQNFLRFGRNAPENDTE------KKRSKRSV---DVQDKR-GSDSFLRFGKA-GD
Aquatica     KYNREMRGNDQNFLRFGRSVD---SN------KKRSKRSV---ETQNKR-GSDNFLRFGKS-GD
Nicrophorus  ---------KGHFLRFGRSGSTVKS--------VRNKRSA--NFEKRPE-RNSNFLRFGR---N
Aleochara    H-S-----KNRDFIRFGRSTRSIKGKPVNKQEKERKKREA----EYYKK-SGSNFVRFGRADRN
Oryctes      Y-S-------------------------------RKKRDTSDVNEEEKR-GNNNFMRFGKRYND
Coccinella   Q-Q--RFSRSRDYLRFGRSPPPPATE------NGRYKRDA--VFDPEVK-RNSNFLRFGR---N
Harmonia     H-Q--RLSRSRDYLRFGRSLPTPGE-------HERSKREA--TFEPDTK-RNSNFLRFGR---N
Dendroctonus Q-R--SARDKSSFLRFGRSISEGSKK-------KRDKRQA-AASADALK-RHENYLRFGRT-AS
Hypothenemus Q-R--STRDKSSFLRFGRSIDEDAKK-------KREKRA----VSETFK-RHENYLRFGRS-SN
Leptinotarsa K-R--THD-KGFHLRFGRSIQEK----------PRSKRQA--TPDKVPK-RTDAYLRFGK---S
Anoplophora  R-R-----DKSAYLRFGKNEMPIFKE-------YRHKRDTFS--DEADK-RSDSFLRFGR---N
Aethina      R-L--HRDRD-THLRFGRSAPVQEQK-------SRAKRDT-AMQEEYKR-GGSNFLRFGR---N
Hycleus      -------DNDNNYIRFGRNVNNEDRK-------HRNKRDASSIYPDYKR-SSSNFLRFGR---N
Tenebrio     R-V--VRDRSGNYLRFGRSVPNTEEK-------VRNKRDT---YSEFKR-GGSNFMRFGR---N
Tribolium    K-A------TTNYLRFGRRN---------------------------KR-DTSNFLRFGR---N

             257
Pogonus      GNFMRFGRMPPIFKNDKDIDAQDD--QNHQNE-------SPVTREKIFPSDQQLSIPANYWPVL
Ignelater    DSFIRYGRNPDGIKINADDTLP---VPTKSN-----FIHGQSAHYPIA-INGRLLHSSPLFSIL
Photinus     DSFIRYGRNQQKTKLNLEEEFP---MPTVSRPLPPSFELEGTLRFPLV-LNDRILHKFPLLTIL
Aquatica     DSFIRYGRNKNEIKKQIDDLYP---LPTVSHPFVNRFGSDPLLYYPMI-FNEKLLHRFPLLKVL
Nicrophorus  DNFMRFGRDPEVVYNQKHNQV---------------------------------------LQML
Aleochara    DNFMRFGRNMNANYLQQPHVAS----ING-----------TDSLCKLLEQMQGRQSQALLLRML
Oryctes      EDYLEDFQGEEAKKQ----------------------------------NLHDKLENAFFLKLL
Coccinella   SNFLRFGRNKDDPMQG----VE----LTQE-------------DLKMLGRLRQ-FYESPLWRLF
Harmonia     SNFLRFGRTKEDHSQH----LE----LTQE-------------DLKLLSQLRQ-FYESPLSRLF
Dendroctonus SNFMRFGRNLGAQGGLSEEELI---RPS-------------------LE-----SRNRHLLDYL
Hypothenemus SNFLRFGRTLR--------------LPS-------------------YE-----SRNKYLINYL
Leptinotarsa SNFMRFGRNHQTTPLSHNNADERLVLVNSAAI--------DHLPPRSFEELKV-PANSPLALLL
Anoplophora  SNFMRFGRDPTNYLANPGNMLS----T---------------------LELLK-MQEQTLANVL
Aethina      SNFLRFGRNPDGVLAMPTSETE----ITHAQ------------INELLGKIKQ-MHDSPLLRLL
Hycleus      SNYLRFGRDNEILGTVPSY----------------------------YNRHQN-LNRGPLVQLL
Tenebrio     SNFLRFGRNNEMTATSD------------------------------PEKVQQ-LQESPLVQLL
Tribolium    SNFLRFGRNNESS------------------------------------------YESPLVQLL

             321
Pogonus      QNFAASLRYTSGKQKCV-----DVTNCPPRQG----
Ignelater    RKIEEEMRQSN-------------------------
Photinus     KKLEEV-RQNH-------------------------
Aquatica     KKLEDEVRQSH-------------------------
Nicrophorus  MDLLRKQKEQ--QKQQQQQQDEDIQA----------
Aleochara    IDAMARNRLQNELQQLQQQQH-GFE--GEFKGHKET
Oryctes      SQISCNMDQ----INCD-----------DRLD----
Coccinella   VQLRLEGQER-----WK-----SDV-----------
Harmonia     AQLEFQGQRR-----CE-----SDV-----------
Dendroctonus KSLLRQSDGRISHLN---------------------
Hypothenemus RNLLRQSLENGSRKN---------------------
Leptinotarsa AQLMARRQEQ--GQRWE-----TFL-----------
Anoplophora  AQLMLHKKEE--NKPCT-----RAA-----------
Aethina      TDLAINARQD--GKKFS-----S-------------
Hycleus      EDLIENLRKE--HGKHA-----DGSA----------
Tenebrio     SELLEHIKKG--QDKNR-----IV------------
Tribolium    SSLLKK---E--ENKQR-----IV------------
```

Figure S30. Alignment of FMRFamide precursors.

```
                   1
Pogonus            MACSFFNVLRFFWLVICIFA-VVVKPDEDPDDSKISMCELSLAQ-LLLLPP--DSMQVQALKRELFRYLEEK
Ignelater          MAYFFSPKILISCGI-LWFT-LLIGIKSEERENSLSACEIEKLLKPILSPPDERTIQAQALRQRLINLLRQG
Ignelater-s.v.     MAYFFSPKILISCGI-LWFT-LLIGIKSEERENSLSACEIEKLLKPILSPPDERTIQAQALRQRLINLLRQG
Photinus           --MRCPPKFLFCCGI-LWFL-LFGEIKSED--TPISSCEIERLLKPILSPPEDRTIQAQALRMRLMNLIRQG
Aquatica           MLCFFPSKILFCCGI-LWLL-FFDAIKSEDPESPLSTCEIERLLKPILSPPDDRTIQAQALRNRLINLIRQG
Nicrophorus        ---MAK------VPI-VLLL-AVFIIRAHCQ----EYMDLDEIFARIFCPEKATTMQAQALRHDIIGKIASK
Aleochara          MG-----TLTVGVVV---LA-LSCA-LAPQLASAQSDEEINSMLEKLFAPESESSMQSRALRREVLRRLYDA
Oryctes            -------------------------INAQEE----CDTNLENMLTSLVSPDEPQTIQLQSLRNELIRRIKEA
Coccinella         MVSGFRKPFAV-----VFLSVFIIMVQSDET----CNMDFERTLEILFAPQEQPSPQIRSLRRYLISIFHNM
Harmonia           MVSRFPLKLLL-----VFLSVYVCMVQSDEP----CNLDFERTLQTLFAPQEQPSPQIRSLRRYFISILHSM
Dendroctonus       MGNLRLINLSV-WCVLVGCALIDNKVKSDNS----CDLDIPESLRILLDPQETDSIQVQALRRQLLRKLQL-
Dendroctonus-s.v.  MGNLRLINLSV-WCVLVGCALIDNKVKSDNS----CDLDIPESLRILLDPQETDSIQVQALRRQLLRKLQL-
Hypothenemus       MGSLGLIQLSM-WCILVGFALIDNKVNSDNS----YDFDTAEALRILLLPEENDSIQVQALRRQLLKQLSNL
Anoplophora        MSFLSQ--TVVSLAA-LVLV-GSYGVNSDNT----CDIDVENTLRTLLSPQEN-SMQVQALRRDVLRKFQEA
Anoplophora-s.v.   MSFLSQ--TVVSLAA-LVLV-GSYGVNSDNT----CDIDVENTLRTLLSPQEN-SMQVQALRRDVLRKFQEA
Leptinotarsa       MAAF--EPIFISVVL-FISA-MLYEVLPDKS----CDIDIEKTLGTLLNPQDNPSIQVRALQIELLKKLEEA
Aethina            ---MRKP-IVGFWCI-LIVD-AVLLQVNSEN----CDGEIENALRKLLSPSEYPTLQLRALRRQLLHKMQQA
Aethina-s.v.       ---MRKP-IVGFWCI-LIVD-AVLLQVNSEN----CDGEIENALRKLLSPSEYPTLQLRALRRQLLHKMQQA
Hycleus            -----MGSFIVRIGV-LMVA-LIVTVKSDD-----YDFDIENALRTILTPQDYPTIQLQALRRSIFNRLLDR
Hycleus-s.v.       -----MGSFIVRIGV-LMVA-LIVTVKSDD-----YDFDIENALRTILTPQDYPTIQLQALRRSIFNRLLDR
Tenebrio           MAVFGAPKFLIGTGI-LMSA-LFFMVKSDET----CDIEIENTLRTLLSPQEFPTIQQQALRKDLLRRFQEA
Tribolium          -MLFGAPKFFFGTGV-LVFA-LIFVVKSDES----CDIEIENTLKTLLTPQDYPSMQQQALRKDLLRRFQQA
Tribolium-s.v.     -MLFGAPKFFFGTGV-LVFA-LIFVVKSDES----CDIEIENTLKTLLTPQDYPSMQQQALRKDLLRRFQQA

                  73
Pogonus            LEEDEL--------------SKRSFASLVRDGNLPYVPKRSIEALARSGYLKSATDDF--------------
Ignelater          FEEQVDNG----------IEEKRSISSLARWNDLPM--KRNLQALARAGYLRSLPDDD----------EIDD
Ignelater-s.v.     FEEQVDNG----------IEEKRSISSLARWNDLPM--KRNLQALARAGYLRSLPDDD----------EIDD
Photinus           LDEQVDNG----------LEEK-------GWDDMAT--KRSLQSLAREGYLRPS--EE----------SADD
Aquatica           LDEQIGNE----------LDDKRSISALARWNDMPM--KRNIQSLAREGYLRSN--ED----------NEDD
Nicrophorus        LAREGINEDEDLVRKVDEMNKKKESYDDDYYRMAAF-GKRNLAALARAGYIRTLPDED-------------N
Aleochara          IEREQLDDDL--------ANYKRSINSLARLGTIPG--KRNLAALAKGGYIRTIPDSSTEQ---------EQ
Oryctes            KQREELENEL--------LQYKRSLSSLARWNNMPG--KRNLESLARAGYIRTLPTTN----------E---
Coccinella         VARAEE---------------------IEGWNTYVY-PKRSLEALARAGYLHTLPDDE-----------DNQ
Harmonia           VARAEE---------------------AEGWNTYVY-PKRSLEALARAGYLHTLPDEE-----------DNQ
Dendroctonus       DCHNL-----ELAELEDAKNY-KSLASLAGWGNIPAEHKRNLEALARAGTWYKNPPEI-----------DQS
Dendroctonus-s.v.  DCHNL-----ELAELEDAKNY-KSLASLAGWGNIPAEHKRNLEALARAGTWYKNPPEI-----------DQS
Hypothenemus       LQPNV-----EFAEQPDDGNYRRTLASLDLWNDLPEEHKRNIEALARSGMACRMFPEL-----------EP-
Anoplophora        LDRLDQEEDE--------RNLKRSLSSLAAWNNLPE-HKRNLEALARAGYLKTLPDED-------------D
Anoplophora-s.v.   LDRLDQEEDE--------RNLKRSLSSLAAWNNLPE-HKRNLEALARAGYLKTLPDED-------------D
Leptinotarsa       AEKQEE------------DENKRALVALAKYNDLPE-HKRNLEALARAGYIKTLPDEE-------------D
Aethina            LEKFEE-EED--------PNLKSSLSALARWNNLPE--KRNLESLARAGYIKTLPDVE-------------D
Aethina-s.v.       LEKFEE-EED--------PNLKSSLSALARWNNLPE--KRNLESLARAGYIKTLPDVE-------------D
Hycleus            FDNGNIDV-----------DIGNDVGVIDRWN------KRNLEALARAGLVRTLPITQQDNNDDNNEHDNDE
Hycleus-s.v.       FDNGNIDV-----------DIGNDVGVIDRWN------KRNLEALARAGLVRTLPITQQDNNDDNNEHDNDE
Tenebrio           LEKANL-DDD--------MNYKRSLSSLAQWDNLPD--KRNLEALARAGYVRTLPSQD-------------D
Tribolium          LDRADD-EDE--------MNYKRSISSLAQWGNLPG--KRNLEALARAGYIRTLPNPD-------------E
Tribolium-s.v.     LDRADD-EDE--------MNYKRSISSLAQWGNLPG--KRNLEALARAGYIRTLPNPD-------------E

                 145
Pogonus            TDDDVKRSIANL--AKNGQLPTFQRPEDEQTQTKRGIESLARNGDLHGK---RK-LENLFENLYGKRNIASL
Ignelater          NDPNLKRSIQYL--VKNGQLPD-YRS----EESKRGIQALARNGELHNT---RQDLEELLDELYEKRNIGSL
Ignelater-s.v.     NDPNLKRSIQYL--VKNGQLPD-YRS----EESKRGIQALARNGELHNT---RQDLEELLDELYEKRNIGSL
Photinus           GD--LKRSIQSL--ARMNQFPV-Q-S----DDLKRSISSLARNGELINN---RREMEELLDEINAKRNIGSL
Aquatica           TDLSLKRSIQSL--ARLNQFPE-YQT----EPPKRSISSLARNGELFDG---RRNLEDIIDELYEKRNIGSL
Nicrophorus        ----GKRSIANL--AKNGQLPN-YQN----DAEKRGIESLARNGELHNK---RE-IEDLIDELYEKRNIANL
Aleochara          EEEQDKRSLESL--VRAGQMPNVAYD----AESKRGIESLARNGELHGK-----------------RNLGSL
Oryctes            --PDYKRNFMTTRSVRNTQSPV--------------------------------------------------
Coccinella         TDSNDKRSLSAL--AKNGQFPLHQFR---------DEESYKRSSGPPNSGDISKEIQKMLDDLYNN------
Harmonia           TDSNDKRSLSAL--AKNGQLPVHRLR---------DEESFKRSGTPVSTNDINKEMQKMLDDLYNN------
Dendroctonus       EDANYKRSMEVL--LKNGPAPLMPES---LENQKRGIESLARNGDFLRR---QP-FQSFLNSQDYKRNIASL
Dendroctonus-s.v.  EDANYKRSMEVL--LKNGPAPLMPES---LENQKRGIESLARNGDFLRR---QP-FQSFLNSQDYKRNIASL
Hypothenemus       QDSTYKRSIEIL--AKNGQHPLVRQP---IENQKRGIEALARNGDLHRP---PN-YQIQQGNMDYKRNLANL
Anoplophora        GDSNYKRSIANL--AKNGQLPMP--S----DGQKRGIESLARNGDLRPK---KD-IQELLDEMYGKRNIGSL
Anoplophora-s.v.   GDSNYKRSIANL--AKNGQLPMP--S----DGQKRGIESLARNGDLRPK---KD-IQELLDEMYGKRNIGSL
Leptinotarsa       ADANYKRSIANL--AKNGQLPTR--K----EDQKRGIESLARNGDLHQN---RE-MLEPQQDAYLA---RSL
Aethina            ANDSFKRSIANL--AKNGQLPHH--------DDKRGIQSLARNGEIHTK---PTSRQDQLDEELYKRNVASL
Aethina-s.v.       ANDSFKRSIANL--AKNGQLPHH--------DDKRGIQSLARNGEIHTK---PTSRQDQLDEELYKRNVASL
Hycleus            DDGNFKRSLSSL--VRNGQLPH---------QNKRGIQSLARNGELIQH---QRNNFDD-------------
Hycleus-s.v.       DDGNFKRSLSSL--VRNGQLPH---------QNKRGIQSLARNGELIQH---QRNNFDD-------------
Tenebrio           EDPNYKRSLATL--AKNGQLPT-YQN----NDSKRGIESLARNGELTTR---RE-IQELLDELYNKRNVGSL
Tribolium          EDPNNKRSLSTL--AKNDQLPTTFQN----NESKRGIESLARNGELHNR---RD-IQELLDELYNKRNVGSL
Tribolium-s.v.     EDPNNKRSLSTL--AKNDQLPTTFQN----NESKRGIESLARNGELHNR---RD-IQELLDELYNKRNVGSL
```

```
                    217
Pogonus             ARGFNYPV-----AG-KRYIGALARGGDLRYSSSDDDFDKRNIASIVRNGKRNVQSLVRTNNLPGGKRYYLF
Ignelater           ARDFNFPS-----YG-KRFLGSLAKSGDLSSRFNG---GKRNLASIMRNGKRNIASLVRSNNLPSGG----Y
Ignelater-s.v.      ARDFNFPS-----YG-KRFLGSLAKSGDLSSRFNG---GKRNLASIMRNGKRNIASLVRSNNLPSGG----Y
Photinus            ARDFNFPS-----YG-KRFLGSLARSGDLS RFES---GKRNIASIVRNGK         RNLPE G----Y
Aquatica            ARDFNFPT-----YG-KRFIGSLARNGDLNGHFSG---GKRNLASVVRNGKHSAATLARTNNLPE-D----Y
Nicrophorus         ARSYSFPY-----G--KRYIGSLARSGELNRF---------------------------------------H
Aleochara           ARSYSLPP-----YG-KRYLGALMKN----------------------------------------------
Oryctes             --DGQFTY-----YD--------------------------------------------------------L
Coccinella          --------------------------------------------------------------IAR------D
Harmonia            --------------------------------------------------------------LAQ------V
Dendroctonus        ARAYSFPVGSTAGFG-KRSIGSIARNGNLPYF----------------------------------------
Dendroctonus-s.v.   ARAYSFPVGSTAGFG-KRSIGSIARNGNLPYF----------------------------------------
Hypothenemus        ARTYNFPVSNGQSYG-KRSLGSLAKNGELPFR----------------------------------------
Anoplophora         ARGFNLPS-----YG-KRSLSSLAKSGDLNYQ---------------------------------------R
Anoplophora-s.v.    ARGFNLPS-----YG-KRSLSSLAKSGDLNYQ---------------------------------------R
Leptinotarsa        VKNYNFPY-----ST KRSLSSLAKSGDLYSR---------------------------------------Q
Aethina             ARSYNFPV-----FNQKRSLSSLARAGDLPN-----------------------------------------
Aethina-s.v.        ARSYNFPV-----FNQKRSLSSLARAGDLPN-----------------------------------------
Hycleus             ----TSDF-----YGSKRNIASLARSGDLNLG----------------------------------------
Hycleus-s.v.        ----TSDF-----YGSKRNIASLARSGDLNLG----------------------------------------
Tenebrio            ARNFNFPT-----YG-KRFLGSLVRNGDSQYS---------------------------------------G
Tribolium           ARNFNFPS-----YG-KRYLASLVRNGELKYS---------------------------------------G
Tribolium-s.v.      ARNFNFPS-----YG-KRYLASLVRNGELKYS---------------------------------------G

                    289
Pogonus             DELKRNIASLARTMGSRYTGKRAVPTAALLRQDGLMKPVDHHHYDDDSSKSSDNDNDRHENDSTDKRNIAAL
Ignelater           DEYKRNIASLARE-GGKFVGKRNV--AALLRQDNYLNERHHQPESEEENQTEK-PDNDLEETENEKRNLASV
Ignelater-s.v.      DEYKRNIASLARE-GGKFVGKRNV--AALLRQDNYLNERHHQPESEEENQTEK-PDNDLEETENEKRNLASV
Photinus            DDYKRNIASLARG-GGKFVGKRNV--AALLRQDNYVNEQNRH-DTDS---THS-PDVEGEEATESKRNLPSI
Aquatica            DEYKRNIASLARD-GDKFVGKRNV--AAMLRQDNYLNEQLRH-QSEDYTHTEK-PVATDEVNNESKRNLASI
Nicrophorus         -NDKRNVASLARG-GNLLYGKRNV--AALLRQDKIHGPNDRS-----YDDMMK-SDAERDSGNGDKRNLASI
Aleochara           --------------SGGFITKRNV--AALLRQDKIHGPNDRSSHMDESTGLGL-SLGKHSVDQGGKRNVGAA
Oryctes             PMYKRNVASLARS-GMKINGKRNV--AALLRQDNYLNSIREGRG---DLQ-----TERPSENYEDKRNIASI
Coccinella          GNTKRNLASIARD--GGFAGKRNA--AALLKNDRYLSHML--------------------NQVDSKRNIASL
Harmonia            ETTKRNIASIARD--GGFLGKRNV--AALLKNDRYLSNML--------------------N   GKRNIASV
Dendroctonus        -YGKRNIQSLARD-G--ALGKRSA---------------------------------DYMERPADKRNIQSI
Dendroctonus-s.v.   -YGKRNIQSLARD-G--ALGKRSA---------------------------------DYMERPADKRNIQSI
Hypothenemus        -YGKRNIQSLARD-G--VIGKRSI---------------------------------DSDDSPLDKRNIQSL
Anoplophora         TQYKRNIASLARD--GKFTGKRAM--PAL-REDEYSVSLH---------------RLPQAEGMEEKRNLQSI
Anoplophora-s.v.    TQYKRNIASLARD--GKFTGKRAM--PAL-REDEYSVSLH---------------RLPQAEGMEEKRNLQSI
Leptinotarsa        SSAKRNIAALARE-GM-IPGKRYG--GSQI----YYNS-----------------EIPFN------------
Aethina             -FYKRNIAALARD-GA--LGKRLE--DSYF---------------------------TEDGTENIKRNIASI
Aethina-s.v.        -FYKRNIAALARD-GA--LGKRLE--DSYF---------------------------TEDGTENIKRNIASI
Hycleus             ---KRNIGALARD-GIRVNGKRNLP---------------------------------------------SI
Hycleus-s.v.        ---KRNIGALARD-GIRVNGKRNLP---------------------------------------------SI
Tenebrio            ---KRNIASLARE-GGRFVGKRNV--AAMLRQDNYLNGQK-----------SN-EKVEGPELDNEKRNLASI
Tribolium           ---KRNIASI--------------------------------------------------------------
Tribolium-s.v.      ---KRNIASI--------------------------------------------------------------

                    361
Pogonus             KA-QMKS--SKFKRST-------ENENETSSTRSKRQADYWD-PSEEYPVPVYQNNNV-FDYEELIHSL-T-
Ignelater           KA-GYK---SKFKRSI-------PYVEMARGGRNKRQADYYDFQNEEYPQPVYQNQNNNMDYEELMHAI-N-
Ignelater-s.v.      KA-GYK---SKFKRSI-------PYVEMARGGRNKRQADYYDFQNEEYPQPVYQNQNNNMDYEELMHAI-N-
Photinus            KA-GYK---PKFKRST-------PYTGDY TLRSKRETDYYDGSNEEYLQPVYQNQNS-REYDEILQAL-A-
Aquatica            KA-GYK---PKYKRSA-------PFTD-----RNKREADYYDVPNEEYIQPVYQNQNN-REYEELLNAL-Y-
Nicrophorus         KA-GYK---QPFKRSV-------ET-------RKKRQADYYE INDEYASPVLQNQNV-FEYEELIKAL-T-
Aleochara           KANGYKP---PSKRSIHLPFLQEPSQEEFYGLKPKRDI---DYFTDEYAMPVYQNQNE-HDYEEMMKAF-T-
Oryctes             KA-QYK---PKFKRSV-------VDD-----KRAKRETDYYDMVNEEYPSPVYQSLFD---YSDD KDL-D-
Coccinella          KA-SYK---PRYKREL-------------------SNADDYE-------DEEYQNIQNVDGYEKLVEELL--
Harmonia            KA-SYK---PRYKREM-------------------NDLDYYE-------DEDYQNLQNIDRYEKLVEEIL--
Dendroctonus        KAQ----------------------------QRNKRQADYFENE------LVYQMPA---DYEDILQDLAAS
Dendroctonus-s.v.   KAQ----------------------------QRNKRQADYFENE------LVYQMPA---DYEDILQDLAAS
Hypothenemus        K------------------------------QRNKRQADYFENE------MVYQIPI--TDYEDYLQEL-SS
Anoplophora         KA-QYK---PKFKRSA-------DETE----KRNKREADYFDVENGEYPSPVYQSPNI-YDYEDWLQDL-T-
Anoplophora-s.v.    KA-QYK---PKFKRSA-------DETE----KRNKREADYFDVENGEYPSPVYQSPNI-YDYEDWLQDL-T-
Leptinotarsa        ------------------------E----------LERMNNHYENGHYFSPNDQNIDY-N----FIQDL-NA
Aethina             KA-QYN---PKFKRSA-------SRT-----------KREIDYDNLEYSSPVYQNTNV-FDYEEMIKEL-T-
Aethina-s.v.        KA-QYN---PKFKRSA-------SRT-----------KREIDYDNLEYSSPVYQNTNV-FDYEEMIKEL-T-
Hycleus             KA-QLLPPNRQFKREL----------------NTNRQKRQIDYDNTEYPSPVYQNQNV-YDYEELMKAL-T-
Hycleus-s.v.        KA-QLLPPNRQFKREL----------------NTNRQKRQIDYDNTEYPSPVYQNQNV-YDYEELMKAL-T-
Tenebrio            KA-QYS---GKFKRAV----------------RSKRQTSYYDDEGGELPSPVYQNQNV-DDYEELVKAL-T-
Tribolium           KA-QYP---G     T----------------RSKRQASYYDGEGGEFSLPVYQNQNV-DDYEELVKAL-A-
Tribolium-s.v.      KA-QYP---G     T----------------RSKRQASYYDGEGGEFSLPVYQNQNV-DDYEELVKAL-A-
```

```
                   433
Pogonus            -GEYPNQEKRFLGAVARQGGWYRSP--------SRFR-PPSKRHIGSLARLGWLPAFRTARRFNRSGRSANE
Ignelater          -GIYPNAEKRFLGSVARS-GWPSSL--------VKYR-NPDKRHIGSLARLGWLPSFRTARRFNRSGRSTNY
Ignelater-s.v.     -GIYPNAEKRFLGRLPQM-GKPKTT--------PTPK-VRRRRISGFY------------------------
Photinus           -EAYPNNDKRFLGSVARS-GWPSSL--------VRYR-NPDKRHIGSLARLGWLPSFRSARRFNRSGRSTNY
Aquatica           -GMYPNNEKRFLGSLARS-GWPTSL--------LKYK-NPDKRHIGSLARLGWLPSFRSARRFNRSGRSTNY
Nicrophorus        -GEYPNTDKRFLGSVAKS-GWFRPSAS------SRFTSRIDKRHIGSLARLGWLPSFRSVRRFNRSGRSSN-
Aleochara          -GEYPA-EKRFLGSVAKS-GWFRPR--------------IDKRHIGSLARLGWLPAFRQIRRTSA-------
Oryctes            -DEYQSFDKRFL---AKS-GWFRQS--------NRFV-PPNKRHIGSLARLGWLPSFRTARRFNRSGRSSSS
Coccinella         -EGQR-AQKRFLGSIAKS-GWYDSPRS----SSMVR-SSPGKRHIGSLARLGWLPTIRNVRRFSRSGRSSS-
Harmonia           -EGQQQPQKRFLGSIARN-GWYNSPRS----SSIRSTSNEGKRHIGSLARLGWLPTIRNVRRFSRSGRSSS-
Dendroctonus       QDAYPVEDKRFLGSVAKS-GWFRPV----SRSYSSWVSLPEKRHVAALARLGWLPTYRSVRRFNRSGRSYLR
Dendroctonus-s.v.  QDAYPVEDKRFLGRIPQM-GKPKTTAIPKSRRYH--------------------------------------
Hypothenemus       QGSYPVEDKRFLGSVAKS-GWFRPT----SQGYSSFVSLPGKRHVAALARLGWLPSYRSLRRFNRSGRSYLV
Anoplophora        -GAYPNAEKRFLGSIART-GWFQPSNS----RYSRFV-SPDKRHIGSLARLGWLPSFRNVRRFNRSGRSTTS
Anoplophora-s.v.   -GAYPNAEKRFLGRLPQM-GKPKTTPA----PKSRRY-H---------------------------------
Leptinotarsa       AQGPSNYEKRFLGSVAKT-GWFQSSLS--SAKDPEFF-KTEKRHIGSLARLGWLPSLRQIRRFNRSGRSTSA
Aethina            -GEYPEPEKRFLGSVART-GWFRPATS----RMSRYN-SPDKRHIGSLARLGWLPIRRYERGFNRSGRSTNL
Aethina-s.v.       -GEYPEPEKRFLGRLPQM-GKPKSTVS----PKVRFP-----------------------------------
Hycleus            -GVYPNTEKRFLGSLARS-GWFRSTAP--STS-RYYQNLPEKRHIGALARLGWLPAFRNVRRFNRSGRSSIY
Hycleus-s.v.       -GVYPNTEKRFLGRLPQM-GKPKTTPP--PKT-RFH------------------------------------
Tenebrio           -GIYPNTDKRFLGSVAKT-GWFRPT--------SRYR-SPEKRHIGALARLGWLPTLRNVRRFNRSGRSTRG
Tribolium          -AAYPNNDKRFLGSVARS-GWFRPN--------SRYR-SPEKRHIGALARLGWLPTLRNVRRFNRSGRSTTL
Tribolium-s.v.     -AAYPNNDKRFLGRLPQM-GKPKTT--------P----SPKTRFH---------------------------

                   505
Pogonus            PVLTEYIDDNVRSYPADGSAEIDTQLSEEPLDEAAKRYLLLPAIDNILLRKLYSNTQNSNQD--LL------
Ignelater          EEC-----GVCRSAEDV-------------------------------------------------------
Ignelater-s.v.     ------------------------------------------------------------------------
Photinus           EEC-----PPCRETTSDGEAEDDPFAQN----AENKRFLLIPAANHLLLRKFPMHTFHN-------------
Aquatica           GEC-----GICRETAADGEAEDDSFSES----PEDKRFLLIPAANSMFLRKLPVRS----------------
Nicrophorus        -EC-------YREASADGQTKEYAFP-EDKLPVDDKRFLLQPAVDRILLQRVLMQPRNH-------------
Aleochara          ---------------PDGQVEDYPVAEDHL-QVIPARHFGPP-T-GLLLDDF-----SDNVEGYIVQRAVQS
Oryctes            CDS-----RF-------------AFPYSR-------------------------------------------
Coccinella         --------RSCREASSDGQNQDYFIT-DNAVPLDNKRFLPVPAMWKVPLQTFNY------------------
Harmonia           --------GRCREASSDGQDEDYFLT-DNT--VDNKRFLPVPAMWKVPLQTFNY------------------
Dendroctonus       SN---TEDIVCRSSQ---------------------------------------------------------
Dendroctonus-s.v.  ------------------------------------------------------------------------
Hypothenemus       TT---GDDNLCRKDSTNGKTEDDSYSKGPTIPLENKRYLLQPAVDNILLRKVFRHPRTIF------------
Anoplophora        DDI-------CREVAPNGQTEDYASAKEPPLSLEDKRYLLQPAVDNILLRKVFMHPRSH-------------
Anoplophora-s.v.   ------------------------------------------------------------------------
Leptinotarsa       ENT-------CRTITSDGQTKVNTNPQHPLITLLDARQKYQPKENVFYLRKMFHHPRSH-------------
Aethina            -DE-------CRALTADGEAEIDSFAKS--------TFPLNIPINNLMVRSHPRHPNAL-------------
Aethina-s.v.       ------------------------------------------------------------------------
Hycleus            NINNIDDNEICR------------------------------------------------------------
Hycleus-s.v.       ------------------------------------------------------------------------
Tenebrio           -EG-------CRETSSDGQAEDDAVA-ENSLSLGDKRFLLQPAVDKILLRKIFMHPRTHPFLSDLS------
Tribolium          -EG-------CREAATDGQTEDDAFA-EDQISLEDKRFLLQPAVDQILLRKIFMHPRTHPAIN---------
Tribolium-s.v.     ------------------------------------------------------------------------

                   577
Pogonus            --
Ignelater          --
Ignelater-s.v.     --
Photinus           --
Aquatica           --
Nicrophorus        --
Aleochara          RA
Oryctes            --
Coccinella         --
Harmonia           --
Dendroctonus       --
Dendroctonus-s.v.  --
Hypothenemus       --
Anoplophora        --
Anoplophora-s.v.   --
Leptinotarsa       --
Aethina            --
Aethina-s.v.       --
Hycleus            --
Hycleus-s.v.       --
Tenebrio           --
Tribolium          --
Tribolium-s.v.     --
```

Figure S32. Alignment of NPLP1 precursors; s.v., splice variant.

```
              1
Pogonus       MMQYAM--------KALAGGLVWAALVSCYCAQATESHDGEHGS------MHMKTPQQDDLQMDSGD-LQMVKRAWN
Ignelater     -MRYAA--------SASVGTLVSAALLACCLKASYTVASSDDPS------MVPKSLED---SNSQLE-TSMSKRDWN
Photinus      -MRYVV--------SASAGSLVWAVVFLFCLKDSYILAASEESP------LSQKSDDD---LNSQLE-SSMLKKDWN
Aquatica      -MRHVV--------SVSVGTLLWTV-LIFSLKARYTCAASEEGL------LGQKPDDD----TSQLE-GSMIKKDWN
Nicrophorus   -MRSAV--------LVIVGAIVCISMLPFSMQAAIDVGSDPDIG------IPK----------ESDE-MQMEKRDWN
Aleochara     -MTYAM--------LSTSRTLLGVAAIFCCLKIGMAVSGNDVGA-------P---KAE---LDPNDQ-MDMEKRGWT
Oryctes       -----MM--------SSNVGKLLWAALLACYLHAGYSIAVSDDSS------LAKS-SDD---TNQQME-SAMNKRDWH
Coccinella    -MRYVF--------ASASHKFLGAAVLAFYLQVNFAYTLPDDAT------ILLKSEDD----NLQTD-TDMVKKNWD
Harmonia      -MRHFF--------ASISHKLLGAALFAFYLQASFAYTLPEEEA------V----DDN----LKQMD-SDMMKRNWN
Dendroctonus  ----MFG-LWPMLRMRICDIFLYSMLIAICLEAAVLA-DV----ANDVPMPPRKSLEE----NPQLE-NDYSKRGWK
Hypothenemus  ----MIG-RWPT-------YFLYVTAYVTCLRAAAISNDV----PMIMAAVPQKSLNE----NPQVD-NDELKRGWK
Anoplophora   -------MRYVPTPTVSLGKMFWAALLACCLQISVVVLADDS--------SAAKSGDE----NLQAEG---------
Leptinotarsa  --------MRYQSTNVNSAKILWATLLACYLQACLTFVVADN--------ADSRGQDD----NLQVDGDEVNKRDWN
Aethina       -MRHVSGVLVAGGQSGGGSIWLWSVVLACCLEAALLPAVVAEDIAPVAPISRSDAIDD----NPQL--NEISKRDWH
Hycleus       -MRDAT-IHSTTTKTASSTRFLCVIFLIYYMEIGLVAALSDE--------IPLQSNND----NPQVD-DEMSKRDWN
Tenebrio      -MKDAV--------AAVAAKMLGLVLLACCLQASITRALSDE--------TPMKSTND----NPQMD-DEMSKRDWN
Tribolium     -MRDAV------------APVLGAVLLTC-YSLQATLALSDE--------TPLKSSND----NPQIE-DEMSKRDWN

              78
Pogonus       SM---PQGAWG--KRAWNNMGATWGKRLA--------N--SYGAADKRAWQKFQGGWGKRWGPEDEW--SLSEN---
Ignelater     R----NLHSWG--KRAWQNLQGGWGKRD-----SSYPL--YYPYADKRAWQNLQGAWGKRLAPEDEY--AMKQLASL
Photinus      R----NLHSWG--KRAWSNLQGGWGKRD-----SSYPL--YYPYNDKRAWQNLQGGWGKRYSSDDEY--AAKLA-AL
Aquatica      R----NLHSWG--KRAWSSLQGGWGKRD-----SS-----YYPYNGKRAWQNLQGGWGKRYAPDDEY--ASRLA-AI
Nicrophorus   R----NLHSWG--KRAWQNLQGGWGKRDLWGPSSSFPL--YV--TEKRAWQNLQGGWGKRFAPEDEY--AMKQLATL
Aleochara     KGNPSMMHSWG--KRAWQNLQGGWGKRDLWGPSNSYPL--YI--TDKRAWQNLQGGWGKRFATEEEY--AAKLAALL
Oryctes       K----SLPLWG--KRAWQSLQGGWGKRDAWDDS-------LYPAAEKRAWQNLQGGWGKRYAPDDIY--TLRLAAAL
Coccinella    K----NLKMWG--KRAWSNLHGGWGIKRSIPEDSEQD----IVIADKRSWENLRSGWGKRNDFMEDV---LSQEPVM
Harmonia      K----NLKMWG--KRAWSNLHGGWGVKRSISDDFGQD----DLVTEKRAWENFRNGWGKRRVPLEDL---TPQELSL
Dendroctonus  T----DLPMWG--KRGWSNLHSGWGKRDSDSDEEL-DG--YPSGMEKRAWQRLQGGWGKRYIPTEEDMV-IRQLA--
Hypothenemus  T----DFPLDGTNKRGWNNLRTNWGKRDIGDDD-L-D------LYQKRSWQKLQGSWGKRTYPSDEDDD-MVHQWPV
Anoplophora   ----------L--KRQWERMQGGWGLKSTLPLNDQIG-Y-STPDREKRAWQNLQGVWGKRFAPSEDDL-TIQELASI
Leptinotarsa  K----ELHMWG--KRGWSSIHGGWALKNNIPSLMNQQAYPPIPIVEKRSWNNLQGGWGKRSAYEEDD--AIEDISSM
Aethina       K----ELKIWG--KRAWNGLHGGWGKRSVPNYDDQMMPYDSPSLVEKRAWNRLQNGWGKRMAPDDEL--AVNQLASM
Hycleus       K----DLHIWG--KRGWNNLHGGWGLKRSIPATTWNE---QATTPDKRAWQNFQNGWGKRFLPNDDEYAASRQLSAL
Tenebrio      K----DLHIWG--KRGWNNLHEGWGRKRSAPS--WGD---Q-PAMEKRAWQNLHSGWGKRFAPEDE-YA-IRQLAAM
Tribolium     K----DLHIWG--KRGWNNLHEGWGRKRSVPA--WEE---Q---QEKRAWQSLQSGWGKRFAPEDE-YA-IRQLAAM

              155
Pogonus       ---VD-TEPQQYDD-YAAV-APS-GGDFEDLIDEDKRAWRAIN-GAWGKRAHGWDSFRGSWGKRD--PAWNNLKGLW
Ignelater     IEQEE-QQQQQEPQ-YSEY----GLGSDSDINTDDKRAWKGLN-GGWGKRSDSWGNFRGSWGKRD--PAWTNLKGIW
Photinus      LE-----QEDQEPQ-YNEF----GNLPETDANDAEKRAWKGLN-GGWGKRSDSWGNFRGSWGKRD--PAWTNLKGIW
Aquatica      LE-----QEEQEPQ-YNEF----GPASDIDLNDGDKRAWKGLS-GGWGKRSDGWGNFRGSWGKRD--PAWTNLKGIW
Nicrophorus   LEQED-QGIQGDAP-YADY----AQITEMDANDDEKRAWKNLNNAGWGKRS-EWGNFRGSWGKRD--PAWTNLKGIW
Aleochara     AD-EE-QLQQQLAD-YNSI-IGEGDAADVENNEEEKRGWRSLNSAGWGKRA-QWGNFRGSWGKRD--PAWTNLKGIW
Oryctes       AEQEE-QEPQLT----SDL----SNDADFEPIDEEKRSWKSLSGTGWGKRS-DWTSFRGAWGKRD--PAWTNLKGMW
Coccinella    TRLE------------TDY----PDSDNLEYNEDEKRSWRQLN-HGWGKRN-KWSEFRGAWGKRRPDPAWNNLKGIW
Harmonia      LENE------------LQY----TDFDNLEENDDEKRSWNQLN-NGWGKRN-KWSEFRGSWGKRSPSPAWNNLKGIW
Dendroctonus  -AALSEDNNENTIDYYP-------DQDFDSSHEVNKRNWRGFS-DGWGKRSDKWDKFRGSWGKRQ--PAWNNLKGLW
Hypothenemus  PDPDSIANDNNVLDYYVYYFNPEQVGDFDGGHEVNKRNWRSFP-EGWGKRSNKWDKFRGSWGKRQ--PAWNNLKGLW
Anoplophora   LEQDS-KEAPP----VDD--------NEF-DIHEDKRSWDKFV-DGWGKRN-KWEKFRGSWGKRE--PAWSNLKGIW
Leptinotarsa  LEPST-RY-------KIPT----DSDDDFIVHDKVKRNWSKFT-DGWGKRS-KWDNFKGTWGKRQ--PAWSNLKGIW
Aethina       FERDP-QMEQP----YTEF----GPEAEIDGIEEEKRNWQQFN-SGWGKRN-NWNKMRGAWGKRE--PAWSNLKGMW
Hycleus       LDLDN-VNQLY----SSEY----GPDFDL-YNDDEKRNWGQFH-GGWGKRS-KWEKFRGSWGKRE--PAWSNLKGIW
Tenebrio      LEPQ-----------YDEY----NPEGDLDVNDDEKRNWGQFH-GGWGKRS-KWDNFRGSWGKRE--PAWSNLKGIW
Tribolium     LDSQ-----------YDDY----NPE--IETNDDEKRNWGQFH-GGWGKRS-KWDNFRGSWGKRE--PAWSNLKGIW

              232
Pogonus       GKRSIQDRISS
Ignelater     GKRNYPDRIAQ
Photinus      GKRSFPDRIAQ
Aquatica      GKRSYTDRITQ
Nicrophorus   GKRSPSERRSQ
Aleochara     GKRSFQDKDAQ
Oryctes       GKRSYSDRIYQ
Coccinella    GKRNYNDNVMI
Harmonia      GKRSYSEKAVL
Dendroctonus  GKRSVSE----
Hypothenemus  GKRSISE----
Anoplophora   GKRSLYQYAN-
Leptinotarsa  GKRSEPEQFLN
Aethina       G----------
Hycleus       GKRSNPDQIAQ
Tenebrio      GKRSVQDQIAQ
Tribolium     GKRSGEK----
```

Figure S33. Alignment of allatostatin B precursors.

```
              1
Pogonus       MR---HSRLICPILMAIF-SLINASENVKRAPSGFQGVRGKKSIQDTYPETAPATGNSNTASSG
Ignelater     --MRRTLL-VISIISAALGGVCAADQPKRRAPSGFTGVRGKKSIPDAAYSDTSQS------L--
Photinus      --MWRIIL-MASLLSASIEKIQTADQVKRRAPSGFTGVRGKKSIPDSTYSDTTQI------L--
Aquatica      --MWKTAL-TISLILATLDNVCTTDQTKRRAPSGFTGVRGKKSIPETAYSDTSQI------I--
Nicrophorus   ---MRVPN-ISIVLATVWCLVSTAAEEQKRGPSGFTGVRGKKSFEDEDFEMRD-----------
Aleochara     ---MRVSH--ITLLLAGICI-VTASESHKRVPSGFTGVRGKKSIPNSEYALIQQE---------
Oryctes       MKYFLFSI-RLYLAIIWTVDIAIAIEQEKHAPPGSAFASETSETTLPARNGNNHI---------
Coccinella    MRFSSTA---TFTFSLMLCL-ARSEDHHKRAPSGFTGVRGKKSIVEDPKGAIFFEDESDDEDGG
Harmonia      MRISSTA---TFTFSLILCL-ARSEDHHKRAPSGFTGVRGKKSIVENAKNAIFFEEDSDEDDAT
Dendroctonus  MKSIRPFT-TWISISLLLSTCCLADEHHKRVPSGFTGVRGKKSIPDEAYSADLDNSNSIEQQTE
Hypothenemus  MKFLHQFTSSITLLILIVSSKAEEEDGTKRAPSAFTEVRGKQFISDEAFNEVLNNKDADQQA--
Anoplophora   ----MTLLSIQILLFTVLCILTTKSEDHHT-FSSLTLVRGRKSRDDSIYDSINKFNYSANIL--
Leptinotarsa  -----------MAFFIIVCLCMMLINATLGTPYGFVAVRGKKADPFYDISRIDMRDLPMELFDE
Oreina        -----------MAFILLLYVCIISFDSIIGTPYGFVAVRGKKANSIYDP-RQDMQELSPEIFDE
Aethina       MQTRRGLP--FLLLLTSLCWCVLAEDHHKRVPSGFTGVRGKKSIPEGAYDEQDAPEASK-----
Hycleus       MHPTTSLN-ITIVMVLICCNLCRAEDHHKRIPTGFTGTRGKKSIQYTTTDETVNN--QFNGI--
Tenebrio      MHSTTIT---TAVVLATIYVVCTAEEHHKRAPSGFTGVRGKKSIPDSAYSTGNAESDATSEL--
Tribolium     MHSTTIT---TAVVLATIYVVCAAEDHHKRAPSGFTGVRGKKSIPDSAYSTGNSDSDSIPEL--

             65
Pogonus       ---------VASAENDILDEMSHA-MEKRAPSAGFHGMRGKKIPITFDNYPYPVMSINDMMLLP
Ignelater     ------------DDLSETAGDGNA-LNKRAP-SGFMGMRGKKPFPD---------PDGFPEDMI
Photinus      ------------NDPAFSSGE-DG-LDKRAP-AGFMGMRGKKPFPE---------SNSLPEDIE
Aquatica      ------------ADPSESV-LNNV-VDKRAP-SGFMGMRGKKLFPD---------SETYNED--
Nicrophorus   -----------------------I-EDKRAP-SGFLGMRGKKPFEG---WT----SLDYPEVGI
Aleochara     -----------------------L-EEKRAP-AGFLGMRGKKPFNS---WL---------EEKR
Oryctes       -----------------DNDNTQY-LAKRVP-SGFFGMRGKKPFEG---WI----SSQEDDIYN
Coccinella    GRGEGQQVAIPEVPYQFAGRPE-GDVSKRAPSMGFVGMRGKKPWAMDSGFY-----EE--DRMP
Harmonia      K-VATNQAPLPVASLQYPSGLV-GDIDKRAPSMGFVGMRGKKPWSLDPRFY-----VEDSDRLP
Dendroctonus  Y--------TPSSDTEAPASFNYI-VEKRAP-SGFFGMRGKKPWNF----L---QSSNYPEELY
Hypothenemus  -------------TVAPEISDNYV-LEKRSP-MGFFGTRGKKPSDW----------LGYNEEMY
Anoplophora   ----------KTKDKPLSVSETLETSVRIAP-SGSITNSENKPFAYMQLWK-----GTYPDMLY
Leptinotarsa  Y-----QSMRPYENKL-------L-AEYLAR----LESFQERQ------WD-----GASTDGQY
Oreina        H-----QDVLPYDNKL-------L-AEYLAK----LDSDQARP------WD-----GF--NAQY
Aethina       -------------TLEEPHPEQQK-VEKRAP-SGFMGMRGKKPYSYGEPWE---GSYQFQDAVY
Hycleus       ------------EDDDDGGNDSLN-IEKRAP-SGFMGMRGKKPFPF---YD---EDNIYPDGIY
Tenebrio      -------------KA-DVVSDVGA-VNKRAP-SGFMGMRGKKPYPV---WE-----GTYPDEVY
Tribolium     -------------KAVDIVSDLGA-VDKRAP-SGFMGMRGKKPFSL---WE-----GTYPDGVF

            129
Pogonus       KRAPMGFMGMRGKKEMEMVG----DEYDKRAPNMGFLGMRGKKMSDED--ESLSDYYMEKRAP-
Ignelater     KRAPSGFLGMRGKKDIDLDS---SSEYDKRAP-SGFMGMRGKKDFEDD--VDTYNSYMDKRAP-
Photinus      KRAPSGFMGMRGKKEN-------LDDYDKRAP-SGFMGMRGKKEYDAD--Y--YGNDFVDKRA-
Aquatica      KRAPSGFMGMRGKKDNDLNN--IVEDYEKRAP-SGFMGMRGKKDFDN--------EYAEKRAP-
Nicrophorus   KRAPSGFQGMRGKKGLDYV-----EVLEKRAP-SGFLGMRGKKEYDDM---------FEKRAP-
Aleochara     PSGPSGFLGMRGKKDDLSED---EGEVYKRAP-NGFFGMRGKKEFEPF--VNE----YEKRAP-
Oryctes       KRAPTGFFGLRGKKDMDYVM-----DFEKRVP-SGFLGLRGKKFYDYMGFDDDETPFYEKRVP-
Coccinella    KRAPNGFFGMRGKKDDDADL---TYDMEKRVH-SGFFGMRGKKDEDPS--EPYSFYMSEKRAPS
Harmonia      KRAPNGFFGMRGKKGDDSDS---AYDMEKRMS-SGFFGMRGKKDDDLS--E-ASLYMSGKRAPS
Dendroctonus  KRAPMGFVGMRGKKDSLEDDEVTWDEPEKRAQ-MGFFGMRGKKLWTDY--------QGEKRAP-
Hypothenemus  KRAPMGFVGMRGKKESLDY---FNEPSLKRAR-MGFFGMRGKRYFVEP------QYQEEKRAP-
Anoplophora   KRARSGFVGMRGKKDSGFVE------F-------------------------------------
Leptinotarsa  KRTPYRLMGVRGKRSASSSDEMNTN---------------------------------------
Oreina        KRTPNGFVGVRGKRAASSLAGINII---------------------------------------
Aethina       KRAPSGFMGMRGKKDNELFG--YD-EYPKRAP-SGFMGMRGKKEYDPF------EYYNEKRAP-
Hycleus       KRAPSGFMGMRGKKDIEFPN--FDEEYGKRAP-SGFMGMRGKKANDQ----DQDNDFYYKRAP-
Tenebrio      KRAPSGFMGMRGKKDMEFTN--YGDEYDKRAP-SGFMGMRGKKEYDSL--SNQYDGYFEKRAP-
Tribolium     KRAPSGFMGMRGKKDMEFAN--YD-EYIKRAP-SGFMGMRGKKDYDSS--SSQ----LDKRAP-
```

```
             193
Pogonus      MGFMGMRGKKEYSP-FM-EEEKRAPADGFFGMRGKKQPARSSFFGMRGKKYPYEFRGKFVGVRG
Ignelater    SGFMGMRGKKTFES-IA-EEMKRAQMSGFFGMRGKKQPSRSSFFGMRGKKYPYEFRGKFVGVRG
Photinus     RGFMGMRGKKSIDE-IN-EEVKRAVLSGFFGMRGKKQPARSSFFGMRGKKYPYEFRGKFVGVRG
Aquatica     NGFMGMRGKKSYED-MV-EEMKRAVMSGFFGMRGKKQPSRSSFFGMRGKKYPYEFRGKFVGVRG
Nicrophorus  SGFLGMRGKKDLETVLL-PEESKRAPNGFFGMRGKKPSRSAGFFGMRGKKYPYEFRGKFVGVRG
Aleochara    SGFLGMRGKKNDE--IDSVELKRAPSSGFVGMRGKKQPGRSGFFGMRGKKYPYEFRGKFVGVRG
Oryctes      SGFFGLRGKREFEDEIE-DDKRAAPENRFFGMRGKKMPARNGFFGMRGKKYPYEFRGKFVGVRG
Coccinella   MGFVGMRGRKSYDDTP--DEFEKRAPSGFFGMRGKKQ---WGQFALRGKKIPYQFRGKFVGVRG
Harmonia     MGFVGMRGKKR-----------------------------WGPFELRGKKIPYQFRGKFVGVRG
Dendroctonus MGFVGMRGKKDFENPIGYYEDIKRAPSGFFGMRGKKHP--SGFFGMRGKKYPYEFRGKFVGVRG
Hypothenemus MGFFGMRGKKDFEDSYY---DNKRAPNGFFGVRGKKQP--YGFFGMRGKKYPYEFRGKFVGVRG
Anoplophora  ---------------------------------GKKQ--SSEFFGMRGKKYPYEFRGKFVGVRG
Leptinotarsa ----------------------------------------------------------------
Oreina       ----------------------------------------------------------------
Aethina      MGFVGMRGKKSYEDEFV---EDKRAPQGFFGMRGKKLQRTSGFFGMRGKKYPYEFRGKFVGVRG
Hycleus      MGFVGMRGKKPT--------DDKRAPSGFFGMRGKKQPRQSGFFGMRGKKFPYELRGKFVGVRG
Tenebrio     MGFVGMRGKKEYD-ELM---EEKRAPSGFFGMRGKKMPRQSGFFGMRGKKYPYEFRGKFVGVRG
Tribolium    MGFMGMRGKKDYD-EIA---DEKRAPSGFFGMRGKKMPRQAGFFGMRGKKYPYQFRGKFVGVRG

             257
Pogonus      KKDSSENAIPEFEYNPSA------DSYATDLDMNDLMMLLSEGQDGP-TWEKRKGPSGFLGMRG
Ignelater    KKSSN-EVGDYGYLNGNLGNDHPLSHLSEGLDLNQLMLLLTENSDA----SLGNG-NEYFEDRV
Photinus     KKSSN-EIGDYGYLNGHLDNEEQLAKFAEDLDLNQLMLLLTDNNGNQ--WD----SSYLEQDRV
Aquatica     KKSSN-EVGDYGYLNGHVDNEMEVPQLTNDFDLNQLLQYLNENNGD---WSHDNANSYAGQEMV
Nicrophorus  KKMSN-EAVPPPEYYEEF----GK--LGQGLDINQLMLLLTENDGYNLAKET---GQDY-----
Aleochara    KKSSE-EAI-------------PMQRLTNDLDFNQLMLLLTENEQYNSPETYYVPENGNSV---
Oryctes      KRLNE-IGAPEYVNSE------EVPRYGGDMDLNQLMLLLNEKSSEQLHD--------------
Coccinella   KKTMSNNGN--------FE-----TEPNYNSDMNTLLMQLLEDEDLH-----------------
Harmonia     KKTSYDSGN--------VE-----TEPYLDSDMNRLLMQLLEDEEMRRTEADEV----------
Dendroctonus KKSSE-MAIPSYYYQDST------TDRLSDPDWNQLMLLLTGRNEFDRSH--------------
Hypothenemus KKSQI-TVPKYIYEEPNV------DKLISDLDYNQLMLLLTEQDESRQ----------------
Anoplophora  KKISHE-LPDKV--NV------DLNSWRKELQSNQLMLVLTEQNEPGGV---------------
Leptinotarsa ----------------------------------------------------------------
Oreina       ----------------------------------------------------------------
Aethina      KKDMT-DAD--LYRNDYL----ARLGNVPNLDMNQLLLLLTEKNDEYEANQIDVADDENRIAEK
Hycleus      KKAPN-QAV------IGE----PI--YEQELDLQQLLYLLTNQPDNGDFWNG--IDENSQ----
Tenebrio     KKASD-NPESDYYNNVDL----NT--LGQDLDLNQLMLLLTENDGASDIWNG--NNEVGQYSQK
Tribolium    KKASP-----DDYYNVDL----DT--VGQELDLNQLMLLLTENEGESDIWNG--NNEVGQYSQK
```

Figure S34. Alignment of Tachykinin precursors.

```
             1
Pogonus      MGNMYF---AVLFLT--IAALCVLSTSLYLEKRVGN-------AANFLGKLRMDPKKRPFCNAF
Ignelater    ---MITSTFFIVAVTVA-MLHYQETRALYLPKR--------ADQFSR---EDIVDPKKRPFCNAF
Photinus     ---MVSSILFVISVSIV-MLH-QDGLALFLPKR--------VDPLTR---TGITDPKKRPFCNAF
Aquatica     ---MVNSTMFLITITII-VLQ-HESSAFFLPKR--------VDPVSR---ADMADPKKRPFCNAF
Nicrophorus  ---MVSTTLFIVVVGF--AFLVIDTESIFLPKR--------ANGYEG--RDSIIDPKKRPFCNAF
Aleochara    MSVSPAFTVIMFVTTV--VVLLADVESVFLPKR--------LNGYDG--KDMVLDPKKRPFCNAF
Oryctes      ---MT--SVFFITILVASSLLLAYTHAVFLPKRDG------VNYLA---KESLIDPKKRPFCNAF
Coccinella   ---MLPTKTIVFS-TIIASVFIGEAYFYVLQKKVSDVFKGGVYTIEERLEDNMDPKKRPFCNAF
Harmonia     ---MLPTKSIVFS-TIIVSVFISHAQLYVMQK------KGGVYTIEERVDDNMNTEKRPFCNAF
Dendroctonus ---MTALTLISVVLYSTAMILIIELDALFIPKR--------MDPVLSARERIILDPKKRPFCNAF
Hypothenemus ---MTALAVLFIALYSTAMTVIMGSEGLFLPKK--------LSPYINTREKIIFEPKKRPFCNAF
Anoplophora  --MTTSSAFFIICACMVAVLLVSEAEAMFLPKR--------GSAPYPTRERVFIEPDKRPFCNAF
Leptinotarsa -MSTVSSMLIVICGCAVVC-LLSEVGSLYLPKK--------GLAPYHTRERVFLEPEKRPFCNAF
Aethina      ---MTTLTLWIICGCVI---YMFQVDAAYLPKR--------GPAPFNTKERI-LDPKKRPFCNAF
Hycleus      ---MTAFRYLIIF---VILAVVIEIDAVFLPRR--------LDFNLK--DK-IVSPKKRPFCNAF
Tenebrio     ---MTTATFFVICIAAV---LTVETRSLFLPKS--------LGQNLAARERV-VEPKKRPFCNAF
Tribolium    MKNMTTAKLFVICIFAA---LAIETHSRFLPKS--------ISKNLGATERV-LEPKKRPFCNAF

            65
Pogonus      TGCGRKRSNLPPLASNLF-------------DQEPPEMENLGTLLDLSAEPAVEDLSRQIMSEA
Ignelater    TGCGRKRSNLPALTNRG----------------EPVEESIGSLLELSAEPAVEDLSRQIMSEA
Photinus     TGCGHKRSNLPALTNRG----------------EPVEESIGSLLELSGEPAVEDLSRQIMSEA
Aquatica     TGCGHKRSNLPALTNRG----------------EPVEETIGSLLELSAEPAVEDLSRQIMSEA
Nicrophorus  TGCGRKRSNLPALSM------------------QGDNDETIDSLLELSAEPAVEDLSRQIMSEA
Aleochara    TGCGRKRSNVPAYPAQP----------------QGYTDENIGSLLELSAEPAVEDLSRQIMSEA
Oryctes      TGCGRKRANIPPLMEIQQQLQQSQQMHRSQSPKHRVLDDSVAGLLELNAEPGIEDLSRQILSEA
Coccinella   TGCGRKRSNLPALTSDG----------------EEIDDSISALLELNAEPAVENLSRQIMSEA
Harmonia     TGCGRKRSNLPALTSDG----------------EEIDDSISALLELNAEPAVENLSRQIMSEA
Dendroctonus TGCGRKRSNLPALFSDG----------------QEMENSINSLLELSAEPSVEDLSRQIMSEA
Hypothenemus TGCGRKRSNLPALSSDG----------------QEVDNSIDSLLELSAEPAVEDLSRQIMSEA
Anoplophora  TGCGKKRSNLPALTDKG----------------EEMDETINSLLELSAEPAIEDLSRQIMSEA
Leptinotarsa TGCGKKRSNLPALADNG----------------EEMDESINSLLELSAEPAVEDLSRQIMSEA
Aethina      TGCGRKRSNLPALSDQG----------------EEIDESIGSLLELSAEPAVEDLSRQIMSEA
Hycleus      TGCGRKRSNLPALTDQG----------------EVIEDSIGSLLELSAEPAIEDLSRQIMSEA
Tenebrio     TGCGRKRSNLPALTDQG----------------EIIDESLGSLLELSAEPAVEDLSRQIMSEA
Tribolium    TGCGRKRSNLPALPEQS----------------EVVDENLGSLLELSAEPAVEDLSRQIMSEA

           129
Pogonus      KLWEAIQEAHREIMRQRQETSDGEEIPAVNPLAGATSGVGVANGKCALPSCIAI
Ignelater    KLWEAIQEANVELARRKQESG-VEN--PDN---EAGTPTRSATASCALPPCYI-
Photinus     KLWEAIQEANVELARRKQESA--ES--LDN---DPSSPARSATASCALPPCYIK
Aquatica     KLWEAIQEANVELARRKQESA--ES--LDN---NPGSAARSASASCTLPPCYIK
Nicrophorus  KLWEAIQEANMEILRRKQESA--EG--YEE---EPATPARSATASCALPPCYI-
Aleochara    KLWEAIQEANMELLRRKQESL--EG--ADD---ELPTPARSATASCALPPCYI-
Oryctes      KLWEAIQEANIELRKEQEAMDN-------Q---ELTQDVRNMPNSCAVPPCYAA
Coccinella   KLWEAIQEANMELNRHRQKTKE-------N---ISQKNKIQVAGTCALPPCYTI
Harmonia     KLWEAIQEANMELNRHRQNSQG-------N---QQQKPGIQGAGTCAIPPCYI-
Dendroctonus KLWEAIQEANLELKKHKNRPMLPSD--FSD---DVSMADRGAISTCALPPCYI-
Hypothenemus KLWEAIQEANLELNRQRQGQPVNKQNKNDE---TSSDSDRGAIATCALPPCYIT
Anoplophora  KLWEAIQEANVELNRRKQQNS-VES--TED---DVAVPAKSATASCALPPCYI-
Leptinotarsa KIWEAIQEANVELNRRRQNPRLLDG--EQN---DAPVPAKSATASCALPPCYI-
Aethina      KLWEAIQEANVELNRRKQGL--SDI--P-E---DDAVPARSATASCALPPCYI-
Hycleus      KLWETIQEANLELNRRKQQSSVENS----D---EDVVPAKSATATCALPPCYI-
Tenebrio     KLWEAIQEANMELHRRRQES--AES--AED---DVAVPPRSAAASCALPPCYI-
Tribolium    KLWEAIQEANMELHRRRQES--AES--SEE---DAAVPARSATASCALPPCYI-
```

Figure S35. Alignment of CCAP precursors.

```
              1
Pogonus       -MHFNMKIFMCTVL-LTIVLGTFMTETEATYRKPPFNGSIFGKR
Ignelater     -MVFAYIRTATFVL-VAVVFISWFMGAEGTYRKPPFNGSIFGKR
Photinus      -MHFSYLKIVTAMT-VVLLVLTLLSMTEATYRKPPFNGSIFGKR
Aquatica      -MNTKFANIMAFIV-LASLVVTFISMVDAAYRKPPFNGSIFGKR
Nicrophorus   -MEAKFAKLFVFVF-LALTMGNLFLDVEATYRKPPFNGSIFGKR
Aleochara     -MSRQFTKVFLVVA-AVLIIAGMLNQVEATYRKPPFNGSIFGKR
Oryctes       -MMTVTSKYFTLFV-IALVLINFMQCAESTYRKPPFNGSIFGKR
Coccinella    ----MFSKSFI-TF-FVLIVAFLVMTADATYRKPPFNGSIFGKR
Harmonia      ----MFSKTTT-FF-FIIFIVFMAMNVESTYRKPPFNGSIFGKR
Dendroctonus  MALKVFYTKLAIVLIASTIL-LMMSPANATFRKPPFNGSIFGKR
Hypothenemus  ---MSFSTKVAILFVAIAFL-LVITPAVAAYRKPPFNGSIFGKR
Anoplophora   -MSGYFGKTVTIFCVAVAFICAIMTVTEATYRKPPFNGSIFGKR
Leptinotarsa  -MQQKFGKVFACLCVAVLILGVLISVSDAAYRRPPFNGSIFGKR
Aethina       ----MNSKATAIVI-FALAIICCLTFSEATYRKPPFNGSIFGKR
Hycleus       ---MFPRKFAICCL-FVVVI--VASLTEATYRKPPFNGSIFGKR
Tenebrio      -MQLGFAKFLTVCI-VATFLASWLVMAEATYRKPPFNGSIFGKR
Tribolium     -MQLALAKVFSVCI-VVIILTSWIEMTEATYRKPPFNGSIFGKR

             45
Pogonus       GS--TVEYDS-ASKALSAMCEIANEACQSWFPSQDKK
Ignelater     GV--TAEYDN-ANKALSALCEIAIDACQPWFPSQEK-
Photinus      GI--TAEYDN-ANKALSALCEIAFDACQPWYPSQEK-
Aquatica      GL--AAEYDN-ANKALSALCEIAFDTCQPWYSSQEK-
Nicrophorus   G---NIEYDS-AGKALSAMCEIASEACQAWFPLQEK-
Aleochara     GL--NQEYDT-AGKTLSAMCEIASEACQAWFPQQEK-
Oryctes       GT--STDYDSGAGKALSSMCEIASEACQAWFPSQDK-
Coccinella    GT--AIEYE--ASKALSAMCEIASEACQAWFPNQERK
Harmonia      GA--AVEYE--ATKALSAMCEIASEACQAWFPIQEKK
Dendroctonus  SGSINTEYDP-AGKALSAMCEIASEACSSWFPSQEK-
Hypothenemus  AGGLSAEYDS-AGKTLSAMCEIANEACQSWFPSQEK-
Anoplophora   TGS-NTEYDS-ASKALSAMCEIANEACQSWFPITQEK
Leptinotarsa  TGT-SNEYDN-AGKALSAMCEIANEACQSWFPTQEK-
Aethina       GS--TNEYDS-ASKALSAMCEIASEACQTWFPTQEK-
Hycleus       AP--NTDYDG-ASKALSAMCEIANEACQSWFPNQDK-
Tenebrio      GA--TNEYDS-ASKALSAMCEIASEACQTWFPTQEK-
Tribolium     GA--TIEYDS-ASKALSAMCEIASEACQTWFPSQEK-
```

Figure S36. Alignment of SIFamide precursors.

```
             1
Pogonus      -MFGCWILIVATVVACVTMS----SGSLARDTWEKPGCHKVGHTRKISIPGCVEFRMTTNACRG
Ignelater    -MLAFWLLLAVT-STNVLLT----STVTAKDAWQRPGCHKVGHTRKISIPNCVEFHMTTNACRG
Photinus     -MLASWLLLAVI-FPNVLLS----STVIARDAWQKPGCHKIGHTRKISIPNCVEFHMTTNACRG
Aquatica     -MQKYWIFLVLI-FLNVLV-----ATVIAKDAWQKPGCHKIGHTRKISIPNCVEFHMTTNACRG
Nicrophorus  MLLVWWYLVAAFSVTDALLLQPMTAAAVAKETWQKPGCHKIGHTRKISIPNCIEIKITTNACRG
Aleochara    -MLTWWFLIAAISLSNAFLS----TSVVARDTWQKPGCHKVGHTRKISIPDCVEFHMTTNACRG
Oryctes      -MLACWLLIAVISLSNALLY----NTVTARDAWEKPGCHKVGHTRKISIPDCVEFPMTTNACRG
Coccinella   -MLVSWFLINLLPILNAFLI----NTAMETDGWRKPGCHKVGHTRKISIPDCVEFKMTTNACRG
Harmonia     -MLISWFLLNLLPLLNAFLI----NTAMETDGWKKPGCHKVGHTRKISIPDCVEFKMTTNACRG
Dendroctonus -MLAYYFLFLVLPFSYGTVLA-----PERDRDWHKPGCHKVGHTGKVNIRDCVSFDITTNACRG
Hypothenemus -MLVYYFLFWLILFSNGGL-------SDTDLDWQKPGCHLVGHKQTINLRGCISFDITLNACRG
Anoplophora  -MLAFWFLVIVVSFSNAFLV----TSVNAKDAWYAPGCHKVGHTRKISIPNCVEFYMTTNACRG
Leptinotarsa -MPPFWFLFVAVSSSIDFIP----GSANAGEAWRAPGCHKVGHTRKISIPNCVEFHMTTNACRG
Aethina      -MLAFWLLMTALSFSNAFLV----SSVNARDAWMKPGCHKVGHTRKISIPDCVEFSMTTNACRG
Hycleus      -MLACWLLFTILSLSHAFLV----TTVNAKDSWQRPGCHKVGHTRKISIPDCVEFHMTTNACRG
Tenebrio     -MLACWLLFTVLSLSDAFMV----TTVNARDAWQKPGCHKVGHTRKISIPECVEFHMTTNACRG
Tribolium    -MLACWLLFTLLSLSDAFMV----KAVTARDAWQKPGCHKVGHTRKISIPECVEFHMTTNACRG

            65
Pogonus      FCESWAIPSGPKA-NPEQPVTSVGQCCNIMDTEDVTARVLCVDGVKMLTFKSATNCACYHCKKD
Ignelater    FCESWAVPSGPKA-SPTQPVTSVGQCCNIMDTEPVEARVLCVDGVRTLTFKSAVTCSCYHCKKD
Photinus     FCESWSVPSGPKT-TPTQPVTSVGQCCNIMDTEPVEARVLCVDGVRTLTFKSAVSCSCYHCKKD
Aquatica     FCESWSVPSGPKA-SPTQPVTSVGQCCNIMDTEPVEARVLCVDGVRTLTFKSAVTCSCYHCKKD
Nicrophorus  YCESWAVPSSTNI-SWTQPVTSVGECCNIMDSELQEVQVFCVEGVRTLTFKSAISCSCYHCRKY
Aleochara    YCESWAVPSGPKA-SPTQPVTSVGQCCNIMDTEPVEARVLCVDGVRTLTFKSAVSCSCYHCKKD
Oryctes      YCDSWAVPSGPKT-SPTQPVTSIGQCCNIMETEPLEARVLCIDGFRTLEFRSATRCSCYYCKKD
Coccinella   YCESWAVPTHHKV-NPAQPITSIGQCCNIMETEPVEARVLCVDGIRTLTFKSATSCSCYHCKKD
Harmonia     YCESWAIPSHQKN-NPAQPITSIGQCCNIMETENVEARVLCADGIRTLTFKSAISCACYHCKKD
Dendroctonus FCESWAIPSDPQNAQISQPVTSIGTCCNIMETEPIEVRVLCKEGIRTLVFKSAVSCACYHCKKD
Hypothenemus YCESYALPSDPRNTQITQPVTSVGTCCNIMESESYEARVLCVDEVRHLVFKSAVTCACYHCKKD
Anoplophora  YCESWAVPSSPKT-TPSQPVTSIGQCCNIMETEPAEVKVLCVDGIRTLTFKSAVTCSCYHCKKD
Leptinotarsa FCESYAVPSLPKI-TPTQPVTSIGQCCNIMETEPVEARVLCVDGMRTLTFKSAVTCSCYHCKKD
Aethina      FCESWAIPSGMKT-SPTQPVTSVGQCCNIMDTEPIEAKVLCIDGVRTLTFKSAVSCSCYHCKKD
Hycleus      FCESWAVPSGPKA-TPNQPVTSVGQCCNIMETEPFEARVLCVDGVRTLTFKSAVSCACYHCKKD
Tenebrio     FCESWAVPSGPKA-TPTQPVTSVGQCCNIMETEPVEARVLCVDGVRTLTFKSAVSCSCYHCKKD
Tribolium    FCESWAVPSGPKA-TPTQPVTSVGQCCNIMETEPVEARVLCVDGVRTLTFKSAVSCSCYHCKKD
```

Figure S37. Alignment of GPA2 precursors.

```
             1
Pogonus      ---MVGYFSWITVAFIC-----I-----------QATIALDSAETLECHRRLYTYRVTQSDDQG
Ignelater    ----MFSLFWIFMAA-TSICG-TQQVIEAGTGP------IDAASTLDCHRRLYTYRVTKADENG
Photinus     ----MFLGIGILLTV-ASISG-TQKIVNADLER------VDPINTLDCHRRLYTYRVTQSDENG
Aquatica     VAAAMLILFGIFLTA-FSICR-TQHIIEAGIET------IDPATTLDCHRRLYTYRVTQADENG
Nicrophorus  MSWMS---AAILALLLMSIVDCQKSLVEADASAGG--TSMESSRTLECHRRLYSYKVTQTDENG
Aleochara    MKVEVILSLLLLAGNVEK-IRAQISLIEAV----PEPINKEMKDPLDCHRSSYTYRVTQSDKNG
Oryctes      ----MF-RFWMSCLALAAVGRCQMNVIDAGL----EPIY---ASTLECHRKIYTYYVSQSDVNG
Coccinella   MQYLISWNICLFMI-TT--SVV--SQIDVDSLIPPEE-------DLECRERVYTFQVTQTDENG
Harmonia     MKYLISWNICFFII-ST--SVDCQSIIDVESLIPPEE-----PESLECRERIYTFQVTQTDING
Dendroctonus ---MFFQ-LVFFLLYIS-TMIDTQSIIEAASFEE-----EVENSTLKCHKRLYNYSVSQVDKNG
Hypothenemus ---MIFQ-LLFFFMYIG-SFVRTQSIIDGIALDN-----VNQKTTLKCHKFLHKHDVTQLDKNG
Anoplophora  ---MINILVWMTLTLCV--VVKGQSIIEANTELQEEEPLKDAAITLECHRRLYTYRVTQTDENG
Leptinotarsa --MMITILLCLAVTL----NSARGVIINADSVLEGEQPLANAGKTLSCHRKLYTYLVTQQDENG
Aethina      ---MSIPPLWLTLGVCS--LVIGQSIIKAPPEF------VEVSETLDCHKRMYTYHVTQTDENG
Hycleus      ---MKSYQFLLIILSNS--LINGQSIIDVGLEP------LDASNTIECHRRLYTYRVTQTDENG
Tenebrio     ---MLSVQVWILVCLSA--LVSSQSIIEAGLEP------LDASGTIECHRRMYTYRVTQTDDNG
Tribolium    ---MLGVQVWLFVGLSA--LVRCQSIIEAGLEP------LDASGTIECHRRMYTYRVTQTDDNG

            65
Pogonus      KQCWDTLSVMSCWGRCDSNEISDWRFPYKRSYHPVCVHAGRSKAVAKLRHCEAGVAPGTDRYVY
Ignelater    KQCWDTLSVLSCWGRCDSNEISDWRFPYKKSSHPVCVHYGRNKAVATLRHCEEGALPSAAKYEY
Photinus     KECWDTLSVLSCWGRCDSNEISDWRFPYKKSLHPVCVHHGRNKALAILRHCEEGALPSAARYEY
Aquatica     KQCWDTLSVLSCWGRCDSNEISDWRFPYKKSLHPVCVHYGRNKVVAILRHCEEGASPSASRYEY
Nicrophorus  KMCWDIIQVWACWGRCDSNEISDWRFPYKKSSHPVCMHYGRNKAVANLRNCEDGALPSAAKYEY
Aleochara    RQCWDTLTVMACWGRCDSNEISDWRFPYKKSSHPVCVHYGRRQSVVELRHCEEGAGEEARRYEY
Oryctes      RQCWDTLSVWACWGRCDSNEIVDWRFPYKKSSHPVCVHYGRTRAVATLRHCEEGAHPSAARYEY
Coccinella   KQCWDALSTVACWGRCDSNEISDWRFPFKKSNHPVCIHYGRNRNVVTLRHCEEGALPEAARYEY
Harmonia     KQCWDALSTIACWGRCDSNEISDWRFPYKKSNHPVCIHYGRNRNVVTLRHCEEGALPEAAHYEY
Dendroctonus KKCWDILSVMACWGRCDSNEIVDFRFPYKKSVHPVCVHYGRNKAFVILRNCEEGALPSAARYEY
Hypothenemus KACWDTLSVMACWGRCDSKEMADYRFPYKKSFHPVCVHYGRRKVFVILRNCQEGALPSAARYEY
Anoplophora  RQCWDTISVMACWGRCDSKEISDWRFPYKKSTHPVCVHYGRNRAFVTLRHCEEGVLKGTDHYEY
Leptinotarsa KQCWDKVSVWACWGRCDSNEISDWRFPFKRSNHPVCVHYGRNRVFVTLRHCEDGIVEGTDHYEY
Aethina      RSCRDKVSVMACWGRCDSNEIADWRFPYKKSNHPVCVHYGRKRSEVILRNCDEGVMPLTRIYQY
Hycleus      KQCWDTLSVWACWGRCDSNEISDWRFPYKKSNHPVCVHYGRNRSIVTLRHCEEGALPTASRYEY
Tenebrio     KQCWDTLSVMACWGRCDSNEISDWRFPYKKSNHPVCVHYGRNRSVVTLRHCEEGADPTTARYEY
Tribolium    KQCWDTLSVMACWGRCDSNEISDWRFPYKKSNHPVCVHYGRNRSVVTLRHCEEGANPSAARYEY

           129
Pogonus      LEAEACRCQPCTSSDTSCEGVRYRGHRNQAMTLLSLDSEDGAFGNDEENIIE
Ignelater    LEAAGCRCQACSSSDTSCEGLRYRPHRSHPDLLGFGPQ--------------
Photinus     LEAAGCRCQACSSSDTSCEGLRYRPHRSQPDLIGFGNM--------------
Aquatica     LEAAGCRCQACSSSDTSCEGLRYRPHRSQPDIYGFGSK--------------
Nicrophorus  MEAGGCRCQFCFSADTSCEGLRYTTHRSH-----------------------
Aleochara    LEAAGCRCQLCSSTDTSCEGLRYRPHRSHPDQLGFRF---------------
Oryctes      LEAAGCQCQLCSSSDTSCEGLTYRSHRSHPDLIGFGLH-K------------
Coccinella   LEAAGCKCQICSSSDTSCEGLRYRPQRSHPQSLGFQLI--------------
Harmonia     LEAAGCKCQICSSSDTSCEGLRYRPQRSHPQSLGFQLS--------------
Dendroctonus YEAADCKCQQCSSLDTSCEGLRYRSQRSEPDR-GFKRIIRGREVDIY-----
Hypothenemus YEAADCKCQQCTSLDTSCEGLRYRAHRSGF--GGFRRLVRGGEEFF------
Anoplophora  LEADDCKCQLCSSSDTSCEGLRYRPHRSHPDSLGFRYNFKK-----------
Leptinotarsa LEADDCKCQVCSSSDTSCEGVRYRPHRSDPDRLGFL----------------
Aethina      LEADECKCQQCSSSDTSCEGITYRPHRSHPDSLGFKIN--------------
Hycleus      LEAAGCKCQLCSSSDTSCEGLRYRPQRSHPGSLGFRIN--------------
Tenebrio     LEAAGCKCQQCSSSDTSCEGLRYRPQRSHPASLGFRIN--------------
Tribolium    LEAAGCKCQQCSSSDTSCEGLRYRPQRSHPASLGFRIN--------------
```

Figure S38. Alignment of GPB5 precursors.

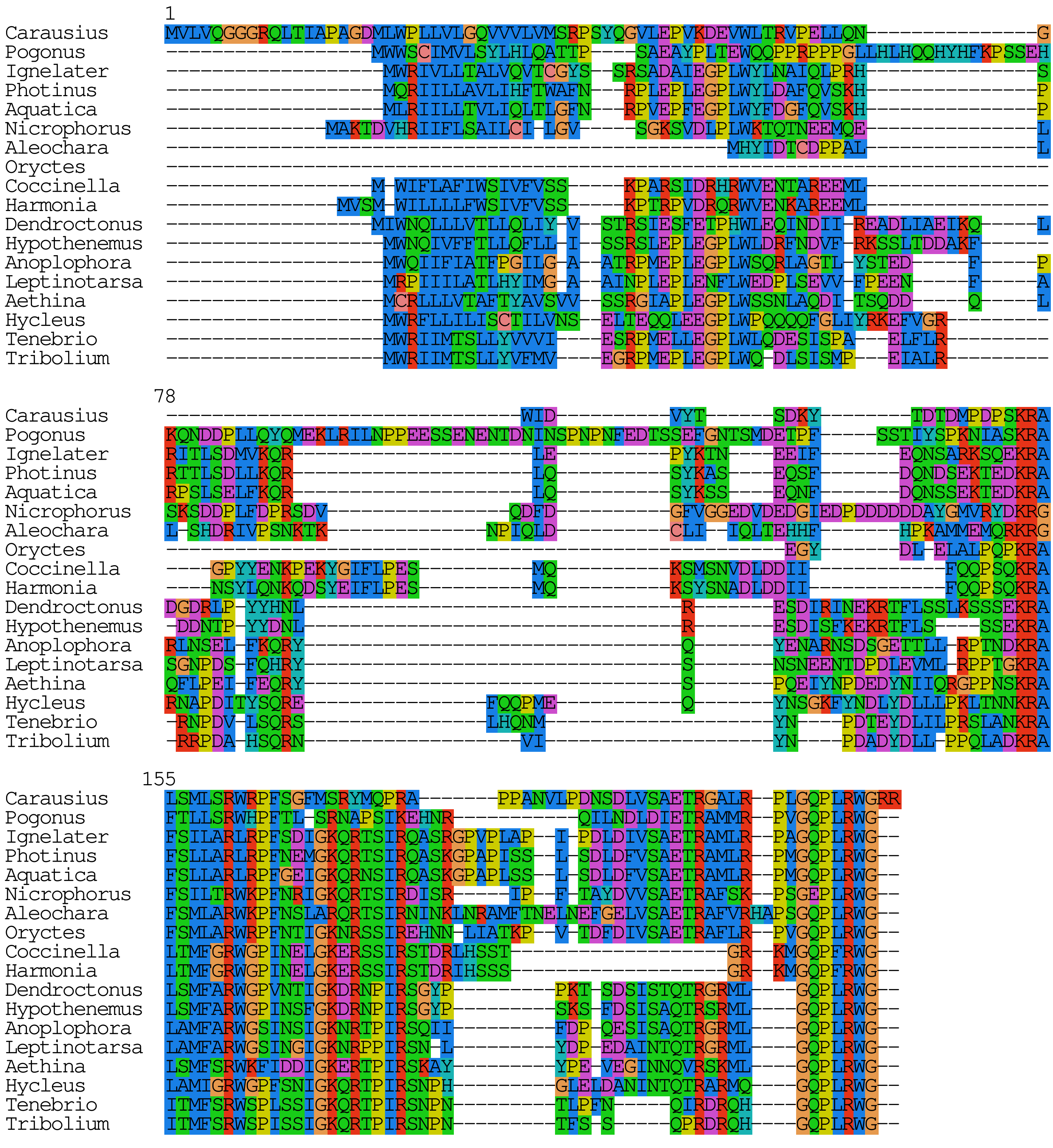


Figure S39. Alignment of Hansolin precursors.

```
                              1
Pogonus                       ---MQKNY---LLVTVVLCLASNSWSAHLI---TPKNCLPCILAANSI
Ignelater                     ----MIIIFALVLVTTVQSYPAGQILY----------------SKDL-
Photinus                      ---MKIITLLIFIVTVTHAYPPGQFLY----------------SDNL-
Aquatica                      ---MKIIIILV-FVTFAYSYPPGQILY----------------SDNL-
Nicrophorus-splice-variant-1  MISLRRQFLVQLLALLVTGQP-----IHLTE--DEMNKDIGIITT---
Nicrophorus-splice-variant-2  MISLRRQFLVQLLALLVTGQP-----IHLTE--DEMNKDIGIITT---
Aleochara                     ------------------------------------------------
Oryctes                       ------------------------------------------------
Coccinella                    ----MIVLLGIILVSRVHCRSFIEGLSEAGVIPQINPTYI-ITSDSM-
Harmonia                      ----MIVLLGIILVLRVDCRSFIQGY------PRVHPGYI-IRSDSI-
Dendroctonus                  ---MNLVVLIIVF---SIGNIRETFARSVVVSSAIGVQDSEAADARIS
Hypothenemus                  ---MILSLFIAIF---SVGHVRQSMAGTLTLVPKHGVADL------LL
Anoplophora                   ---MVLVLVAII-FLALCGGLVKSFPARLAMEAGPGE---RVFQKGLS
Leptinotarsa                  ----------------------------KVEATTLGE---QDFEK---
Aethina                       ---MIVILVGLICVAGTYGAIAT----DFD---AYFNTMSQPRKGDLG
Hycleus                       ----MRITLV-ILILVICGSLFNNITTALP---TIFNTLP--------
Tenebrio                      ----MRIAFGVIFVTGIFGGFFGDNASASP---VHHHHVI---SKDLD
Tribolium                     ----MRIAFGVIFVTGIFGGFFAENVFALP---VARHHVI---TKDLD
Tribolium-splice-variant      ----MRIAFGVIFVTGIFGGFFAENVFALP---VARHHVI---TKDLD

                              49
Pogonus                       KRSEENAMDSLLEVANEEEDDSL----------ERRYILDMDKIHEQA
Ignelater                     SNSVDEVYGSYFTLSDNRKDNNEVNNHPYSQPYLNKMIIPP--PSPQQ
Photinus                      SNSVDEVYDSYLSVSDDDKDD---VNPPPNS-LLNQLIENE----N--
Aquatica                      SNSVDEIYDTYLSVADDDKED---INPPPPEPYLNKLIEET--INL--
Nicrophorus-splice-variant-1  TSSGNQIRNSILQYSDEKIADELQ----YVSQSINRLYSEESYPWNG-
Nicrophorus-splice-variant-2  TSSGNQIRNSILQYSDEKIADELQ----YVSQSINRLYSEESYPWNG-
Aleochara                     ---GEQ-------------------------QRMNSLYRNL-LIFNR-
Oryctes                       ----------------DDEDYNNNILHHKFYAKLQRYYLDK--LQNQQ
Coccinella                    -DKINEEYDTYIDLKDNDAESNNF----DFISEEERFFSNG--RQF--
Harmonia                      -GKIKEEYDAYIDLKDNEDDSNEY----DILSEEERFISNG--HQF--
Dendroctonus                  QNDQKQISGNYISYLDNQA----------------ALLVDQ--IQQYK
Hypothenemus                  PLEPTSFGKNHLFIFNQQEPSRIQNDIAYFDKREGQMNVKK--LNNYR
Anoplophora                   SDSVNEVYDSYYAISKKNEENEET----NVIARVQRVDLNK--IKYSH
Leptinotarsa                  --KPAIILNDYYLFDN---NQEEL----DVFSQIHQINSHN--FRLSH
Aethina                       KTTPDRIQETYSLANRE-EIREE--------KTTSSLFLNK--LKEVY
Hycleus                       ----KTNNELYLSVSDKYSDNSNRLD--KMSARRRQVTLHQ--PKFLQ
Tenebrio                      DTTADKMNKPYISISHDRADNSDK---ADVITRVQKVFVDS--PKSNV
Tribolium                     L---DEMYKSYTSISNDRDDYNDK---V-----TSRVYLDS--PQISV
Tribolium-splice-variant      L---DEMYKSYTSISNDRDDYNDK---V-----TSRVYLDS--PQISV

                              97
Pogonus                       RAAAFDALIKLHDQSDNVAIIHRHKKQRHDKRFLNCKFATLPVRHARA
Ignelater                     SEDLAKSEP--------------ESRRVERTAY-------LH------
Photinus                      --ANLISPA--------------ETQRVEQTSI-------VH------
Aquatica                      --ESLINPE--------------DAQRVERTAF-------VH------
Nicrophorus-splice-variant-1  ------------------------VSPF-KPEV-------AP------
Nicrophorus-splice-variant-2  ------------------------VSPF-KPEV-------AP------
Aleochara                     ------------------------VADMLENDY-------LQ------
Oryctes                       A---ITNLE--------------D-SEIQRTAY-------LL------
Coccinella                    -------GR--------------ERDREWIQTY-------IL------
Harmonia                      -------GT--------------DRDRELIQTY-------IL------
Dendroctonus                  N----KELS--------------KEVQSRIEEF-------IM------
Hypothenemus                  -TIAMEPLP----------A---LVRDRIEDSF-------V-------
Anoplophora                   ------------------------IYDINGDTY-------IL------
Leptinotarsa                  ------------------------FYNLRGRTV-------LV------
Aethina                       K---FDSLG--------------RPQKIEERAY-------LL------
Hycleus                       -----HALK--------------PKKLDEETAY-------LV------
Tenebrio                      ----SKNGK--------------Q-QKTKQTAY-------LL------
Tribolium                     ----SKNGK--------------Q-QKTKQTAY-------LL------
Tribolium-splice-variant      ----SKND----------------------------------------
```

```
                              145
Pogonus                       AEAREGQKGGKRRAAVSDLSLCHFKICNMGRKRN--QRFIHSFNEYDN
Ignelater                     ---IQTK-QKDKRYA-SYLTLCHFKICNMGRKRT--TRYFHVISSLDN
Photinus                      ---VQTR-EKNKRYS-SYLTLCHFKICNMGRKRT--TRYFQVMKTLDN
Aquatica                      ---VQTQ-EKNKRYS-SYLTLCHFKICNMGRKRT--TRYFQMMRSLDN
Nicrophorus-splice-variant-1  ---GKIRRQRKKRFLVTDLTLCHFKLCNMGKRKT--TRTFQKISRASS
Nicrophorus-splice-variant-2  ---GKIRRQRKKRYL----TLCHFKLCNMGKRKT--TRTFQKISRASS
Aleochara                     ---KHRNKRNDPKSYVSDAAPCYFKLCNLGKRRA--------------
Oryctes                       ---IKTMKQRDKRYTSSYLTLCHFKICNMGRKRT--TRYFHMIKSLHD
Coccinella                    ---VKTMRQRNKRYT-SYLTLCHFKICNIGKKRR--TRYIHGMQSEKG
Harmonia                      ---VKTLRQRNKRYS-SYLTLCHFKICNMGKKRR--TRFVHGMESSRE
Dendroctonus                  ---LKQQSNNQRQKRRPRLTPCLFKICNMGRKRQ--HHVLCTYKS---
Hypothenemus                  ---FLKMKTTRERSKRSHLMPCYYKICNMGRKRL--------------
Anoplophora                   ---VKAFRHRNKRFT-SYHTLCPFKICNMGRKRTRTRRYIEPIRPVGD
Leptinotarsa                  ---LKTLKHRNKRYT-NYLTLCPFKICNMGRKRMRAKRSNNKIKRSSD
Aethina                       ---VKTLKHRNKRYS-SYLTLCHFKICNMGRKRN--TRYKSFNDMEND
Hycleus                       ---VNRM-QRDKRYI-SYLQLCHFKICNMGRKRS--ARYFHMIRKLDD
Tenebrio                      ---VKTMRQRNKRYI-SYLTLCHFKICNMGRKRT--TRYFHMIRRLDN
Tribolium                     ---VNTMSQRNKRYI-SYLTLCHFKICNMGRKRT--SRYFHMIRQLDD
Tribolium-splice-variant      ------------------LTLCHFKICNMGRKRT--SRYFHMIRQLDD

                              193
Pogonus                       -----
Ignelater                     GT---
Photinus                      KT---
Aquatica                      KT---
Nicrophorus-splice-variant-1  PNWRI
Nicrophorus-splice-variant-2  PNWRI
Aleochara                     -----
Oryctes                       DHT--
Coccinella                    V----
Harmonia                      NRI--
Dendroctonus                  D----
Hypothenemus                  -----
Anoplophora                   NDET-
Leptinotarsa                  GRYSS
Aethina                       LEYYK
Hycleus                       NES--
Tenebrio                      ES---
Tribolium                     NEA--
Tribolium-splice-variant      NEA--
```

Figure S40. Alignment of CNMamide precursors.

```
                 1
Pogonus          -MRAVVVATVLCLLCSGVRVNAWGGLFNRFSPEMLANMGYGAHGGFAQPFIQRAGGEENILESFDEG--EGDDEPCY
Ignelater        -MKGLIVLIAVCVLG-K-QADAWGGLFNRFSPEMLANMGYGGHSGF------IQRAGDEGILEEFEGE---GDDEPCY
Photinus         -MKSVIVIIAISVLT-Q-RAHAWGGLFNRFSPEMLANMGYGGHSGF------IQRAGDEGILEEFETD---SDDDPCY
Aquatica         -MKAFVIIVAVCLLA-H-RAYAWGGLFNRFSPEMLANMGYGGHSGF------IQRAGDEGILEEFEGE---AEDEPCY
Nicrophorus      -MKGLIALLAICLLG-R-QTEAWGGLFNRFSPEMLANMGYGGHGGGGG--GFRTGDEGILEEFEVD---SDDDPCY
Aleochara        -MKVIIALLALCVLG-R-QTEAWGGLFNRFSPEMLANMGYGAHGG---G--LPRGADEGILEEFEAE---EDADSCY
Oryctes          -MKGLIAMVAICLLG-K-YADAWGGLFNRFSPEMLANMGYGGHGNSIA---RIGGGDEGILEEFEIN--RGDEDPCY
Coccinella       -MRSLISLLAICLVG-QQTAHAWGGLFNRFSPEMLANMGYGGHG-GFIQ--RAGEGDEGILEEYSSD--V-DEEPCY
Harmonia         -MKSLITLLALCLVG-QQTAYAWGGLFNRFSPEMLANMGYGGHG-GYMQ--RSGEGDEGILEEYTND--V-EDEPCY
Dendroctonus     -MKSLAALLAVFLVC--HQVHAWGGLFNRFSPEMLANMGYGGHGGGMM-----RDGDDNFLEEYVNEAEEEGREPCY
Hypothenemus     -MKSFAALMAVLLVV--NRVSAWGGLFNRFSPEMLANMGYGGHGAGIL-----RDEENFLDEQYAN---VEDDGPCY
Anoplophora      -MRAAIAILAFCLLG-Q-KAHGWGGLFNRFSPEMLANMGYGGHGGGFIS--RNGEGDEGIMEEFVSE--NSEDEPCY
Leptinotarsa     MMKTIVAILAFCYVG-R-EVNAWGGLFNRFSPEMLANMGYGGHGGGGFS--R-AEGDDGILEEYVSE--NGDDEPCY
Aethina          -MPTLVTILALCLVG-R-QAHAWGGLFNRFSPEMLQNLGYGGHGG-F----ARGDGDEGILEEFAKE--G-DMDPCY
HycleusITG       -MRTLILLFVVCTMS-F-ETHAWGGLFNRFSPEMLANMGYGGHS-GFIQ--RTGDGDEGILEEYASD--N-DDEPCY
Tenebrio         -MRALIILFMACLLG-H-KAHGWGGLFNRFSPEMLANMGYGGHG-GFIQ--RTGEGDEGILEEYASE--G-DEEPCY
Tribolium        -MRALIILFTACLLG-H-KAHAWGGLFNRFSPEMLANMGYGGHG-GFIQ--RTGDEDKGILEEYTNE--G-EEEPCY

                78
Pogonus          GKRCTANEHCCPGSVCVDVDGVIGSCLFAYGRRAGELCRRDSDCESGLLCAEADQGA-ARVCRPPVHQDKQYNEPCH
Ignelater        GKRCTANEHCCPGSVCVDVDGVVGTCLFAYGRRAGELCRRDSDCESGLLCAEPESGGSARVCRPPVRQDKQYSEPCN
Photinus         GRRCTANEHCCPGLVCVDVDG-AGTCLFAYGRRAGELCRRDSDCESGLLCTESESGGATRICRPPVRQDKQYSETCN
Aquatica         GRRCTANEHCCPGSVCVDVDGVVGTCLFAYGRRAGELCRRDSDCESGLLCAEPETGGSTRVCRPPVRQDKQYSETCN
Nicrophorus      RKSCTANEHCCPGQVCVDVDGVVGTCLFAYGRRVGELCRRDSDCESGLVCTESEQTSSSRICRPPVQQDKQYSEPCN
Aleochara        GRKCSANEFCCPGQICIDVDGVYGQCLFTYGRRVGELCRRDSDCESGLVCAEAEQGISTRVCRPPV-----------
Oryctes          ERPCTANEHCCPAQVCIDVDGAVGTCVYAYGRRIGELCRRDSDCESGLVCSEGDQGISARICRPPVQQDKQYSELCN
Coccinella       GKRCTANEHCCPGSVCVDVDGVVGSCLFAYGRRVGELCRRDTDCESGLVCAEAEPGTMTRVCRPPVHQEKQYSELCN
Harmonia         GKRCTANEHCCPGSVCVDVDG-VGSCLFAYGRRVGELCRRDTDCESGLVCAEAEPGTMTRVCRPPVHQEKQYSELCN
Dendroctonus     GKKCTSNEQCCPGLVCVDTETLVGSCLFAYGRRVGELCMKDSDCESGLVCAEADMGPNARVCRPPVRQDKQYSEPCH
Hypothenemus     GKKCTANEHCCPNSVCVDVDGVVGSCLMTYGRRVGELCRSDSDCESGLVCAEADSGPSARICRH---QDKQYSEPCH
Anoplophora      GRRCTANEHCCPGSVCVDVDGVMGSCLFAYGRRVGELCRRDSDCESGLVCAEADGGVSTRVCRPPVHQDKQYSEPCN
Leptinotarsa     GKRCTANEHCCPGSVCVDVDGVIGSCLFAYGRRLGELCRRDSDCESGLVCAEAEVGASTRVCRPPVHQDKQYSETCN
Aethina          GKPCTANEHCCPGSVCVDVDGVVGSCVFAYGRRVGELCRRDSDCESGLVCAEAEPDVPTRVCRPPVHQDKQYSETCN
HycleusITG       GKRCTANEHCCPGSVCVDVDGVVGSCLFAYGRRVGELCRRDSDCESGLVCAEAEQGVS-RVCRPPVHQDKQYSEPCN
Tenebrio         GKRCTANEHCCPGSVCVDVDGVVGSCLFAYGRRVGELCRRDSDCESGLVCAEAEPGVSTRVCRPPVHQDKQYSEPCN
Tribolium        GKPCTANEHCCPGSVCVDVDGVVGSCLFAYGRRVGELCRRDSDCESGLVCAEAEPGVSTRVCRPPVHQDKQYSEPCN

               155
Pogonus          MSGECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVASDQIKNSVQHTAGEKRLTG---YKRPMH--
Ignelater        MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVASDQIKNSIQHTAGEKRLTGFTGFKRPMH--
Photinus         MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVASDQIKNSIQHTAGEKRLTGFTAYKRPMH--
Aquatica         MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVASDQIKNSIQHTAGEKRLTGFTAYKRPMH--
Nicrophorus      MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVAADQIKNAIQHTAGEKRLTGLAAFKRPMH--
Aleochara        -SSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVASDQIKNSIQHTAGEKRLTGLTNYKRPMH--
Oryctes          MSSECDISRGLCCQLQRRHRQQPRKVCSYFKDPLVCIGPVAADQVKNAVQHTAGEKRLTRFTAFKRQMH--
Coccinella       MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVATDQIKNSVQHTAGEKRLTGFSSFKRPLPMH
Harmonia         MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVATDQIKNSIQHTAGEKRLTGFTSFKRPLPMH
Dendroctonus     MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVATDQIKNAIEHTAGEKRITGLA-FKRPIMH-
Hypothenemus     MSSECDISRGLCCQLQRRHRQVPRKVCSYFKDPLVCIGPVATDQIKNIVEHTAGEKRITGMA-YKRSMMH-
Anoplophora      MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVASDQIKNAIQHTAGEKRLTGLTSYKRPMH--
Leptinotarsa     MSSECDISRGLCCQLQRRHRQVPRKVCLYFKDPLVCIGPVAPDQIKSNIQHTAGEKRLTGLSTFKRPMH--
Aethina          MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVAIDQIKNAVQHTAGEKRITGMA-YKRPMH--
HycleusITG       MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVASDQIKSAVQHTAGEKRLTGLATFKRPMH--
Tenebrio         MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVASDQIKNTIQHTAGEKRLTGLASFKRPMH--
Tribolium        MSSECDISRGLCCQLQRRHRQAPRKVCSYFKDPLVCIGPVASDQIKSNIQHTAGEKRLTGLAGFKRPMH--
```

Figure S41. Alignment of ITG-like precursors.

```
               1
Pogonus        ----------MAITRISVKIFILVSNL--FLLFTLASSIRNQRWEDIMRWDGTSN-----TIFDVPDSK--------
Ignelater      -------------KTSLLLLYLVLTSC--IMQICKSQKNRSKRWDLDGDFE-TVKNYE--TFKSFEDAVLVPERTLC
Photinus       MN-------ILKIKNVTHPLFLITVLC--LMW-IHCQKIEFPEWDTKRDVQ----YFD--TFM---------EKPLC
Aquatica       MN-------IWQPFKITK-ILLITICC--FIRLGNGQKFDLSEWETDRNSE-PTKELD--TLL---------NKSLC
Nicrophorus    MGSWTKGHQFIATCTTMFKSTVTFIVI-LCVVSIGANRFWEKS--DLMKWNEKTKNLP-----NFSQDC--------
Aleochara      ----------------MFKFCIILISFFYFINEITSENLWNKE--ETLKNIQTSRNIH-----SDEKNF--------
Oryctes        ----------------MWLLLNAAIVC---FSITKSEILKAFPPTENMDFHPEKSNPK-----RFPE----------
Coccinella     ------------MK--LH--VSLIVLS--FFHILFASDAWKESQENGIDYTLDN-------------TE--------
Harmonia       ------------MK--LH--ASLLVIS--LFHILFALDSWKESQENGGDYSFDN-------------GE--------
Dendroctonus   MTFRYRIAYLMLLAHLSKFKLFYVVAC---R-----EDSRISNFDDNF-----------------PQ----TD---C
Hypothenemus   ---------MWVLTKWNAYFV-FIVFC---I-----EFAASITWLDED-----------------PV----NNHGDV
Anoplophora    ----------------MILNVITLIVC--SVLYG-ESMGWKERNVGNTVFSESESEIDKATCLDAPQEF--------
Leptinotarsa   ----------MKKKKKEFFTILLLVTT--QIQYCFTSERWNKIEN---L-------------MDYDDNE--------
Aethina        ---------------------MLLTME--VH----THTILPENSETEWEYDDML-------------NN--------
Hycleus        -----------------------------------------------------------------------------
Tenebrio       ------------MKNLII--FLFLIST--FFTLNKSMEIMKNRNYRLLDYDEMN-------------NS--------
Tribolium      ------------MKKIII--AILLTLT--FIKFTKPMDIWKDKNYNFLDYDEVD-------------DR--------

              78
Pogonus        --EQKILDDYDCSTIGQCDLDKYNKESIKMIETNELPLKRMKSNTGLSSWRTFDKLLQIDDSKPGAASGIVPYFHTS
Ignelater      SG-NSLEDCENDRDN----------------LMYSMPLRKHQQT-------HNAPVSSTFITK------KTAPIFQS
Photinus       AI-NSVNTCEDERNV----------------AFFNWLFKNRKE--------NSM-TTQPFSTNKFSKSIRTVPLF-Q
Aquatica       SSTNFLESCEDNRDA----------------FLYRLMLKKRKDR-------NTA-LTKAFTLNKLTKSIRTPPTL-F
Nicrophorus    --DDSLGDC-PNEISHMEF-------------------KRKQ---------NPKTLVLSDLPREDPMFKYLPTSQ--
Aleochara      --IDN--YC-KENNCNYEIE------------------KRKD---------NHISLEN--FPLDESKFTFFPAFHHH
Oryctes        -----------IRTMGE-----------------DYPFYTH----------LFDNEIVKEINTKKS-PFITLKSIRS
Coccinella     --NDYLEEFSDNNLFGERQLDKYSKST--------------R---------KNRNEMQQLAPNELK-SKKTVFYQSS
Harmonia       --NDNLEEFSGNDIFGESQMSNYAKSV--------------R---------NNRNEIQKLIPNEMK-SKKTAFYQSS
Dendroctonus   SN-EVNSKCYRNY---DTD----------LANLFNFAVDRRRS----------------------K-NYKSKSKRIS
Hypothenemus   QS-VEMSQCISNGKFREGC----------FSDSFNFFFKNKLKN------------M---KRTIKK-KSSISSLYDP
Anoplophora    --CKNSQDI-SNSLTGHFLKKKA---------------------------SAYSLSVSIFSGMPGN-SKLFPVFQS-
Leptinotarsa   --TKKVEDI-SRSNIARVIPK-------------------------------FFKKMYPMVNDFLK-TKSRPLFETP
Aethina        --LHKI--------QHRRQDDS-------------------D---------LEADFVKKSANYPES-KTRKIQFFDA
Hycleus        --------------------------------------------------------------MEHE-EQFVPLYHTT
Tenebrio       --EE--SKCVDNDLCRN------------------------------------------------------------
Tribolium      --CD-------NEICQNNFDDLIKRKV--------------K---------DNEDMTYQSDVIGKK-TTRLSPYYHP

             155
Pogonus        LVPLPCSCGSHYRLINLGRAYVPRYLVTAVCDADACGATGAYKCRQRSYTVRVLKKKDAGNAHPPELEPDQVNNPPL
Ignelater      HPGLACRCGGEHTYKDLGLHYYPRYVPTVICKPGGCYA-G-FQCRPKKYNITVLRKKGIEEV-----VGKESNSESF
Photinus       KKGLSCACKGEPEMKYLGIMYFPRQLPTIKCKPSSCLG-GPYQCRPKEYKVHVLKQKQAEDL-----DVSDATCSST
Aquatica       SKGFSCSCVMEPYLQDLGIMYFPRHIPTVICKPGSCFG-GPYQCRPKHYKIQVLKQKQFDDL-----DREETSSGIV
Nicrophorus    PEENQCSCGRENRFVDLGPNTYPRYIADMVCNAGGCG--I-YSCRPKTYKLWVMKRRTSGAS-------AFSDLDPI
Aleochara      QTIRQCSCYHEKRMIDLGPEYFPRYVPDMMCLSGSCG--PNFSCKPRRYKVWVMRRRQDDE---------EMGMEPI
Oryctes        QNYYTCSCEMDYQYTDLGADYT--FLYTVSCKGGRCAL---GFCRPINYKIPVLRRRKNEE---------DANSSSL
Coccinella     HNIHPCSCGMEYRLLDLGHLYFPRQIHSMICKEGSCG--NLYRCIERYYKVRVLKK-NPTE-------KSGRLSSIL
Harmonia       HNITPCSCGMEYKVLDLGHLYFPRYIHSMICKDGTCR--NPYRCSERYYKVRVLKK-NPHE-------RSGRPSSML
Dendroctonus   STPFPCPCEVIFGLVDFGNSIYPRYAHSGICKPGTCG--DRHLCQPKGYKITVLRRRDSKDELS--EDFRLLKNPNL
Hypothenemus   NLNVPCQCEMSYRLMDLGSSRYPRYIHTGVCKKNSCG--LFHRCQAKKYKIRVLKKRDPDTEFL------ETESQHL
Anoplophora    FHQCPCSCKMDYPLLDLGRKYFPRFLQTGVCKTDSCG--RHQRCVEKKYKVVVIKQKDQDVP----------ETRGL
Leptinotarsa   TIMGKCSCELDFQPVDLGRFYFPRFIQTGSCQADSCE--RHYSCIERKYVVQVLKYKDPKTV-------DETSSVTL
Aethina        IQPHTCSCGMEFRLLDLGPTYFPRYVPTGVCNRNTCG--PMKECVARQYKIRILKEKDAN--------TDTPNEIFI
Hycleus        TSLTFCGCNLTVKMRDLGLGFYPRFLKDGKCNPRSCK--DGFSCREKHYKVRVLKHRSTPFPT---AADIQSSDSAL
Tenebrio       ---------------TFGEQVKRRYLHTGVCKSEICR--GLYRCVERHYKVRVLKQRDPRS-------PEIKTTMTL
Tribolium      SRPMPCSCGIEFRVLDLGHQYYPRYLHSGVCKSELCG--GPYRCIERHYKVRVLKQKDPRN-------PEIRPSMAL

             232
Pogonus        PESLRKEWKLETFTVTVACECGAEKI
Ignelater      PELLKEFFIPEIHTVAVACECTP---
Photinus       PHSVRDFFIPEEITITVACECAP---
Aquatica       PHSLKDTFISKTFTITVACECLP---
Nicrophorus    PMELRANWSGLGVLVTVDCECGL---
Aleochara      PKELREFWIGENVSATVDCECS----
Oryctes        PSELSENFVREMKSITIGCQCKP---
Coccinella     PGNLRQHWTPEIVDVVVGCECKP---
Harmonia       PANLRQYWTSETIDVVVGCECIP---
Dendroctonus   PKSLKENWVAYLYNIVIDCKCIPFEK
Hypothenemus   PESLAENWMPDFVNIVIGCECVPNTK
Anoplophora    PSSLQDSWVSESVSVSIACECVPI--
Leptinotarsa   PKLLRDSWISESVPVTVACDCVREFK
Aethina        PKSLSEYWTVEFVTVTVSCECHFQI-
Hycleus        PEGLKDYWKAASVPITVACECL----
Tenebrio       PDTLKGMWQPELVTVTVACECSL---
Tribolium      PDTLKGTWLSETITVTVACECSV---
```

Figure S42. Alignment of PTTH precursors.

```
              1
Pogonus       MPQTQSSSKSAINSTSCVSTRTTRRLRTGNVRCAAVWLAAACALALGGNGGSGTGGSRLAIPSG
Ignelater     ----------------------MHHRSSTCVSTRGVWVCMALALVLQLAQ--------------
Photinus      ----------------------MHRRSSTSIYTKGVWVCMALTLVIQLAHC-------------
Aquatica      ----------------------MHRRSSTSLNTKGVWVCMALTLVFQLTQ--------------
Nicrophorus   -----------------------MHHRSASISTQRVWAFMALALVLQMVS--------------
Aleochara     -----------------------MHQRSTSIGTQGVWAYMAVALMLQMTAC-------------
Oryctes       -----------------------MNHRSTSVSTQFVWVCMALTLVVQIAL--------------
Coccinella    -----------------------MNYLSSKVSTTLLWVCMALVLFLSGSF--------------
Harmonia      -----------------------MNYLSSKVSTTLLWVCMAFIFFFGSSL--------------
Dendroctonus  -----------------MSQRSSQLEQRGRGTRRAAVWCLAIALVVQSARA-------------
Hypothenemus  -----------------MSQRSSQLEQRGRGTRRAAIWCLAIALVVQSARA-------------
Anoplophora   ---------------------MSHRSLKSVTNTQAVWVCMALALIFHITS--------------
Leptinotarsa  ---------------------MSNRSSSSVRGTQVAWWCMAIALILHVSS--------------
Aethina       ---------------------MSHPLSQSVRSTQIVGVCMALALVVQVVV--------------
HycleusITP    --------------MFQIPEKHMNCQSPKSISRQAIWSCMALAVLLIQVTT-------------
Tenebrio      ----------------------MSYRSSIIVNTQAVWVCMTLAVIIQAVTS-------------
Tribolium     ----------------------MNYRSSKSISTQAVWVCMVLAVVFQEITS-------------
Tribolium     ----------------------MNYRSSKSISTQAVWVCMVLAVVFQEITS-------------

             65
Pogonus       ATASPAGERT---VAVPPVHHLTK-RSFFDIQCKGVYDKSIFARLDRICEDCYNLFREPQLHSL
Ignelater     ---SSPTQRS---RPLL-SHHLTK-RAFADQQCKGVYDKSIYARLDRICEDCYNLFREPELHTL
Photinus      ----TPTQRS---RPLL-SHHLTK-RAFADQQCKGVYDKSIYARLDRICEDCYNLFREPELHSL
Aquatica      ---STPTQRS---RPLL-SHHLTK-RAFADQQCKGVYDKSIYARLDRICEDCYNLFREPEIHGL
Nicrophorus   ---GSPAQRM---SPLL-SHHLSKRGGFLDLQCKGVYESSIFAKLDRICEDCYNLFREPKLQGL
Aleochara     --DCSPAER----KPLL-THHLTK-RSFYDIQCKGVYDKSIFARLDRICEDCYNLFRDPHLHSL
Oryctes       ---SSPLQRT---HPLL-THHLMKRKIFFQLECKGQYNKSRFARLDRICDDCYHLFREPEVDRM
Coccinella    ---GSPANRS---SNLL-SHHLAK-RSFFDIQCKGVYDKSIFARLDRICEDCYNLFREPQVHSL
Harmonia      ---GNPTNRS---SGLL-SHHLAK-RSFFDIQCKGVYDKSIFARLDRICEDCYNLFREPQVHSL
Dendroctonus  ---GSPWRRGS--GPLL-THHLSK-RSFFDIQCKGVYDKTIFAKLDRICEDCYNLFREPQVHSL
Hypothenemus  ---GSPWRRGSNNGPLL-THHLSK-RSFADIQCKGVYNKQIFARLDRICEDCYNLFREPHILSL
Anoplophora   ---GSPANRS----PLL-THHLTK-RSFFDIQCKGVYDKSIFARLDRICEDCYNLFREPSIHSL
Leptinotarsa  ---GSPANRS---PELLSRHHLAK-RSFFDLQCRGVYDKSIFARLDRICVDCYNLFREPSVHSL
Aethina       ---GSPS-RS----PLL-THHLTK-RSFHAIECQGVYDKSIFARLDRICEDCYSLFREPEVHSL
HycleusITP    ---SPLMDRS----PSLLAHHLTK-RSFFDIQCKGVYDKSIFARLDRICEDCYNLFREPQLHSL
Tenebrio      ----IPTNGS----PVLLPHHFTK-RSFFDIQCKGVYDKSIFAKLDSICEDCYNLFREPQLHSL
Tribolium     ----SPAGRS----PAFLPHHFTK-RSFFDIQCKGVYDKSIFAKLDSICEDCYMLFREPQLHNL
Tribolium     ----SPAGRS----PAFLPHHFTK-RSFFDIQCKGVYDKSIFAKLDSICEDCYMLFREPQLHNL

            129
Pogonus       CRKNCFTTDYFKGCLEALQLTDEMEQIQTWIKQLHGAEPGV---------------
Ignelater     CKKSCFTTDYFKACLESLQLQDEMEQIQTWIKQLHGAEPGPV--------------
Photinus      CKKNCFTSEFFKACLDSLQLQDEMEQIQIWIKQLHGAEPGV---------------
Aquatica      CKEECFTNDYFKACLDTLQLQDEIEQIQIWIKQLHGAEPGV---------------
Nicrophorus   CKNNCFTTNIFKGCLESLQLNDEMEQVQTWIKQLHGAEPGF---------------
Aleochara     CRKNCFTTDYFKGCLEALQLTDEMEQVQTWIKQLHGAEPAV---------------
Oryctes       CRKNCFTTAIFKGCLEALQLGDELDEIQASIKELHGAEARV---------------
Coccinella    CRRNCFTTDYFKGCLDTLQLSDELQQIQTWIKQLHGADPGV---------------
Harmonia      CRKNCFTTDYFKGCLDTLQLSDELQQIQTWIKQLHGADPGV---------------
Dendroctonus  CMKNCFTTAYFKGCLETLQLTDEMEQVKDWIKQIHGAPPDE---------------
Hypothenemus  CMSNCFTTSYFNGCVESLLKLDEMPTFRKMIEYLAK--------------------
Anoplophora   CRQDCFTTKFFKGCIDTLQLSDEMEQIKSWIKQLHGADPGV---------------
Leptinotarsa  CRDNCFTSDYFKGCVDTLQLSDHIPEIMDMIKQLKGAKA-----------------
Aethina       CRKDCFTTSYFKGCLDTLQLQDEMEQVQTWIKQLHGADPKL---------------
HycleusITP    CRKDCFTTDYFKGCLDTLQLSDEEQQIQTWIKQLHGADPGV---------------
Tenebrio      CRKNCFTTDYFKGCLETLQLSDEEAQIQLWIKQIRGAELGGLGPSVSPPNTS----
Tribolium     CRKNCFTTDYFKGCIDTLQRSDEEAQIQLWIKQIRGAELGGLGTLIAPFLFSNTLT
Tribolium     CRKNCFTTDYFKGCIDTLQRSDEEAQIQLWIKQIRGAELGGLGPSASPPNTS----
```

Figure S43. Alignment of ITP-A precursors.

```
Pogonus       MPQTQSSSKSAINSTSCVSTRTTRRLRTGNVRCAAVWLAAACALALGGNGGSGTGGSRLAIPSG
Ignelater     ----------------------MHHRSSTCVSTRGVWVCMALALVLQLAQS-------------
Photinus      ----------------------MHRRSSTSIYTKGVWVCMALTLVIQLAHC-------------
Aquatica      ----------------------MHRRSSTSLNTKGVWVCMALTLVFQLTQS-------------
Nicrophorus   -----------------------MHHRSASISTQRVWAFMALALVLQMVSG-------------
Aleochara     -----------------------MHQRSTSIGTQGVWAYMAVALMLQMTAC-------------
Oryctes       -----------------------MNHRSTSVSTQFVWVCMALTLVVQIALS-------------
Coccinella    -----------------------MNYLSSKVSTTLLWVCMALVLFLSGSFG-------------
Harmonia      -----------------------MNYLSSKVSTTLLWVCMAFIFFFGSSLG-------------
Dendroctonus  -----------------MSQRSSQLEQRGRGTRRAAVWCLAIALVVQSARAG------------
Hypothenemus  -----------------MSQRSSQLEQRGRGTRRAAIWCLAIALVVQSARAG------------
Anoplophora   ---------------------MSHRSLKSVTNTQAVWVCMALALIFHITSG-------------
Leptinotarsa  ---------------------MSNRSSSSVRGTQVAWWCMAIALILHVSSG-------------
Aethina       ---------------------MSHPLSQSVRSTQIVGVCMALALVVQVVVG-------------
Hycleus       -------------MFQIPEKHMNCQS-PKSISRQAIWSCMALAVLLIQVTT-------------
Tenebrio      ---------------------MSYRS-SIIVNTQAVWVCMTLAVIIQAVTS-------------
Tribolium     ---------------------MNYRS-SKSISTQAVWVCMVLAVVFQEITS-------------

             65
Pogonus       ATASPAGERT---VAVPPVHHLTKRS-FFDIQCKGVYDKSIFARLDRICEDCYNLFREPQLHSL
Ignelater     ----SPTQRS---RPLL-SHHLTKRA-FADQQCKGVYDKSIYARLDRICEDCYNLFREPELHTL
Photinus      ----TPTQRS---RPLL-SHHLTKRA-FADQQCKGVYDKSIYARLDRICEDCYNLFREPELHSL
Aquatica      ----TPTQRS---RPLL-SHHLTKRA-FADQQCKGVYDKSIYARLDRICEDCYNLFREPEIHGL
Nicrophorus   ----SPAQRM---SPLL-SHHLSKRGGFLDLQCKGVYESSIFAKLDRICEDCYNLFREPKLQGL
Aleochara     --DCSPAERK----PLL-THHLTKRS-FYDIQCKGVYDKSIFARLDRICEDCYNLFRDPHLHSL
Oryctes       ----SPLQRT---HPLL-THHLMKRKIFFQLECKGQYNKSRFARLDRICDDCYHLFREPEVDRM
Coccinella    ----SPANRS---SNLL-SHHLAKRS-FFDIQCKGVYDKSIFARLDRICEDCYNLFREPQVHSL
Harmonia      ----NPTNRS---SGLL-SHHLAKRS-FFDIQCKGVYDKSIFARLDRICEDCYNLFREPQVHSL
Dendroctonus  ----SPWRRGS--GPLL-THHLSKRS-FFDIQCKGVYDKTIFAKLDRICEDCYNLFREPQVHSL
Hypothenemus  ----SPWRRGSNNGPLL-THHLSKRS-FADIQCKGVYNKQIFARLDRICEDCYNLFREPHILSL
Anoplophora   ----SPANRS---PLL--THHLTKRS-FFDIQCKGVYDKSIFARLDRICEDCYNLFREPSIHSL
Leptinotarsa  ----SPANRS---PELLSRHHLAKRS-FFDLQCRGVYDKSIFARLDRICVDCYNLFREPSVHSL
Aethina       ----SP-SRS---PLL--THHLTKRS-FHAIECQGVYDKSIFARLDRICEDCYSLFREPEVHSL
Hycleus       --ASPLMDRS---PSLL-AHHLTKRS-FFDIQCKGVYDKSIFARLDRICEDCYNLFREPQLHSL
Tenebrio      ----IPTNGS---PVLL-PHHFTKRS-FFDIQCKGVYDKSIFAKLDSICEDCYNLFREPQLHSL
Tribolium     ----SPAGRS---PAFL-PHHFTKRS-FFDIQCKGVYDKSIFAKLDSICEDCYMLFREPQLHNL

            129
Pogonus       CRSQCFSTQYFKGCLESLLLT--EEEQKYHQMIEYLGKRSN--
Ignelater     CKKNCFKTDYFVACLEVLELE--EEREEIVKSVKQLNHRRSRH
Photinus      CKKNCFKSPYFQTCIDTLNLG-SEAAREVYEWVKRMNKRRL--
Aquatica      CKKNCFKTPNFQACIDAFNLG-DDEKKNFYSLVKQVNRRRL--
Nicrophorus   CKSDCFSSDYFKGCLEALLLN--EEMDKFAKMVEYIGKK----
Aleochara     CRSECFSKSYFRGCVEALLLS--EEWEHLNRSVHYLGRK----
Oryctes       CRLNCFKSAYFKGCLKALLIDSVDEIKSIEEDIEITGK-----
Coccinella    CRNRCFSTRIFKGCVDSLLLS--EELPKFSKMIEYLGK-----
Harmonia      CRNRCFSTRIFKGCVDSLLLS--EELPKFSKMIEYLGK-----
Dendroctonus  CMSKCFTTEYFTGCVESLLKS--DEMPSYRKMIEYLAK-----
Hypothenemus  CMKDCFTTEYFKGCLETLQLT--DEMEQVKEWIKHIHGASPDK
Anoplophora   CRSECFSTKYFKGCVESLLMV--EDWPKFEKMIDYLAK-----
Leptinotarsa  CRSQCFSTKIFKGCVESLLEM--EEWSKLKKMIDFVGK-----
Aethina       CRNKCFSTKYFQGCVDSLLLT--DEMPEFNRTIEYLSGY----
Hycleus       CRSQCFTTKYFKGCVESLLLN--EEMPKFLKMIEYLAK-----
Tenebrio      CRSQCFSTKYFVGCVESLLLS--EEMPNFRKMIEYLSK-----
Tribolium     CRSECFSTKYFVGCVESLLLS--EEMPKFRKMIEYLSK-----
```

Figure S44. Alignment of ITP-B precursors.