

Supplemental Materials

Supplemental Figure S1. Protein alignment of GHF 9 enzymes from insects.

Supplemental Figure S2. Phylogenetic tree for amino acid sequences of NIEG1 and reported insect endogenous endo- β -1,4-glucanases.

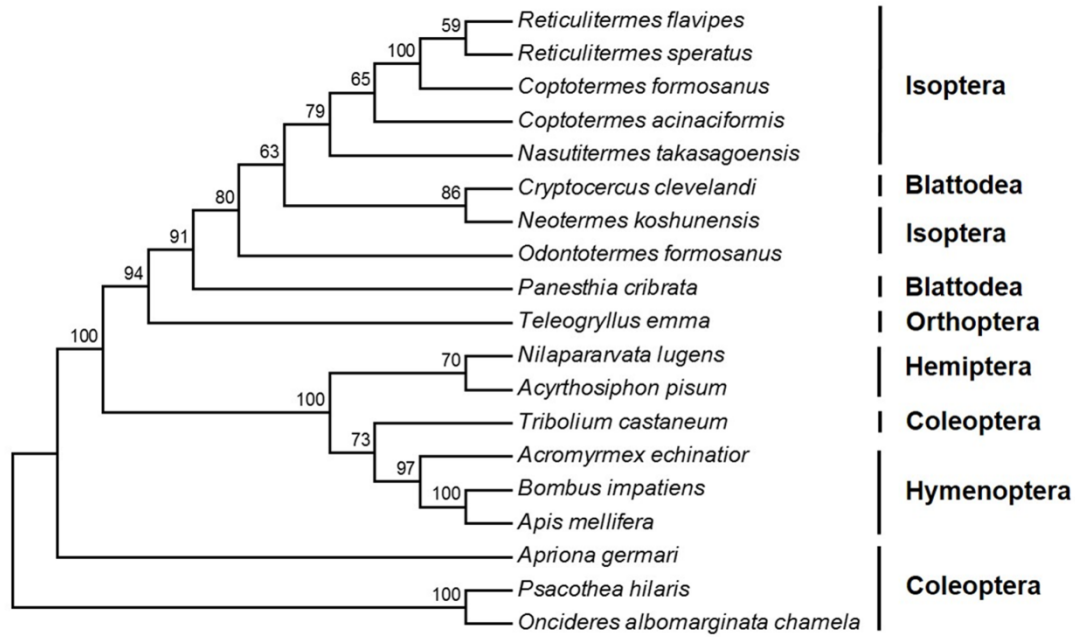
Supplemental Figure S3. The growth phenotype of BPH nymphs 6 days after they had been injected with either *GFP* (left) or *NIEG1* (right) dsRNA. Representative photographs are shown.

Supplemental Table S1. Primers and probes used for qRT-PCR and PCR.

ApM WYATNMNFVILAS.....FLLAGVARCVDYDEVLRISLIFYEACRSGLFVNNRIF	53
Bi	MELNAFVITYVASLAIASIAEIDTINANFFYVVKPIEEDENDYARVLEISLIFYEACRSGLFENNRFIF	67
AeM M YRQILFAILITSTCEINAVPTEDAKAIENGNDYACVLEISLIFYEACRSGLFKNNRFIS	61
CaM R V F V C L I S A L A I C Q A A Y D Y T V I L K S S L I F Y E A C R S G L P A D C K V T	46
TcM F Y S L W L L F I T E G S A Y D Y R H A L K I S L I F Y E A C R S G L P A S N R F I T	45
MbM W V F A Y L L S A L A I C Q A A Y D Y K T V L S N S L I F Y E A C R S G L P A D C K V T	46
OfM R V F V C L I S A L A I C Q G A Y D Y K T V I R N S L I F Y E A C R S G L P A D C K V T	46
NLM N F S H L G L Y V L L A T S T F T Y C V F G S K Y D Y S K V V H I S L I F Y E A C R S G L P A N N R I F	54
◆ ◆		
Ap	WRPDSATTDFKQNGEDISGGYYLACDFVKFCFTMASTTTTLAWGYLSYKDAYEEVNEWENALGSIWF	120
Bi	WRPDSALDRGLNGEDLTGGYYLACDFVKFCFTMASTTTTLAWGAVSWPEAYNAAGQLIEELRKAIKW	134
Ae	WRGDSALYDRGNSGEDIIGYYLACDFVKFCFTMASTTTTLAWGTVNWPEAYIAAGQLIEIRRAIKW	128
Ca	WRKDSALDRGNNGEDLTGGYYLACDFVKFCFTMASTTTTLAWGLVLYEAGYSSAGATLDGRKAVKW	113
Tc	WRPDSALDRGLNGEDLTGGYYLACDFVKFCFTMASTTTTLAWGMLFRHAYNEAGCYCHGLDAIKW	112
Mb	WRKDSALNRKQNGEDIIGYYLACDFVKFCFTMASTTTTLAWGLVLYEAGYSSAGALIDGRKALKW	113
Of	WRKDSALNRKQNGEDIIGYYLACDFVKFCFTMASTTTTLAWGLVLYAAGYTSAGALIDGRKAVKW	113
NL	WRGDSALNRGLKEDIIGYYLACDFVKFCFTMASTTTTLAWGCLSYKDAYKAAGEWKEVLNAIKW	121
Ap	AAEYLIKCHVSDYEFYQVGGFFSDGGEWGRPEDMNMSRFSKIDKNRPGS DLAGE TSAALSASVMV	187
Bi	ATDYLIKCHVSEYVYQVGGFFSDTEWGRPEELNTRPAYKIDPEHPGS DLAGE TAALAAASSMV	201
Ae	STTYLIKCHVSENVLYQVGGFFSDHMEWGRPEELNTRPAYKIDAEHPGS DLAGE TAALAAASSIV	195
Ca	ATDYLIKCHVSEYVYQVGGFFSDHAYWGRPEDMTARPAYKIDAEHPGS DLAGE TAALAAASSIV	180
Tc	ATDYLIKCHVSEYVYQVGGFFSDHAFWGRPEENMTRPAYKIDIEHPGS DLAGE TAALAAASSIL	179
Mb	ATDYLIKCHVSEYVYQVGGFFSDHSYWGRPEDMTISRPAEKIDTSKPGS DLAGE TAALAAASSIV	180
Of	ATDYLIKCHVSEYVYQVGGFFSDHSYWGRPEDMTISRPAEKIDAEHPGS DLAGE TAALAAASSIV	180
NL	ATDYLIKCHVSEYVYQVGGFFSDHMEWGRPEDMNMSRFAKIDAEHPGS DLAGE TAALAAASSMV	188
Ap	FTKILSNFSASCLSHARQLYMFATCYRGLYHEATKGAACQYBSTTYGDELTWAAAWLWKATKESQYL	254
Bi	ERNHIFPEYSAKCLRHAKELKFAEYRGLYHEATKGAACQYBSTDYCEDELAWAAVWLKATNLTITYL	268
Ae	FSSVISEMSAKCLRHAKELKFAEYRGLYHEATKGAACQYBSTDYCEDELAWAAVWLKATGRTKYF	262
Ca	EKGVLESSYNNLLRHARQLYMFATCYRGLYHEATKGAACQYBSTDYCEDELAWAAVWLKATNENTYL	247
Tc	FENENCSYSDELLRHATELYDFATNYRGLYHEATKGAACQYBSTDYCEDELAWAAVWLKATKIVKYI	246
Mb	EKNVLESSYNNLLRHARQLYMFATCYRGLYHEATKGAACQYBSTDYCEDELAWAAVWLKATNENTYL	247
Of	EKAVLPSYNNLLRHARQLYMFATCYRGLYHEATKGAACQYBSTDYCEDELAWAAVWLKATNLTITYL	247
NL	EKEETPEMSDCLLRHARQLYMFATCYRGLYHEATKGAACQYBSTDYCEDELAWAAVWLKATGENNYI	255
Ap	DDAEYMYMKVFLKERENEFYFNKRVAGVQVLLAELTNCSETEAIQNFCDYNIYVQKTPKGLIYIE	321
Bi	EDAEHHYCHEHLKERENEFYFNKRVAGVQVLLAELTNCSETEAIQNFCDYNIYVQKTPKGLIYID	335
Ae	DDAEHHYCHEHLKERENAFYFNKRVAGVQVLLAELTNCSETEAIQNFCDYNIYVQKTPKGLIYIE	329
Ca	TKAEISLYNERGLGNWGAENWENKVSQVQVLLAKITSKQAKETVQCYVDYLINNCQKTPKGLIYID	314
Tc	EQAEITFYTKFIFKRENEFFYFNKRVAGVQVLLAELTNCSETEAIQNFCDYNIYVQKTPKGLIYIE	311
Mb	NTAEISLYNERGLGNWGAENWENKVSQVQVLLAKITNKCCYKDTIKCYVDYLINTCKKTPKGLIYID	314
Of	NTAEISLHNCFCIQWNGGFSWDAKVSQVQVLLAKITNKCCYKDAIKCYVDYLINACQKTPKGLIYIE	314
NL	DEAEYFYMRFLKRENEFFYFNKRVAGVQVLEAQLTKRDDYVCAARAFCDYNIYVQKTPKGLIYIE	322
Ap	KSGILCHAAANVFLICLCAAL.IGIM.SAQCFAFAFYCILDYLGDDGGSYVVGFGKNEYIKCFEASS	386
Bi	KEGILCHAAANVAVFLICLCAAL.IGIM.SAQCFAFAFYCILDYLGDDGGSYVVGFGKNEYIKCFEASS	402
Ae	KSGILSHVAVNVAFLICLCAAL.IGIM.SAQCFAFAFYCILDYLGDDGGSYVVGFGKNEYIKCFEASS	396
Ca	QWGLIRHAAANVFLICLCAAL.IGIM.SAQCFAFAFYCILDYLGDDGGSYVVGFGKNEYIKCFEASS	379
Tc	KSGILSHVAVNVAFLICLCAAL.IGIM.SAQCFAFAFYCILDYLGDDGGSYVVGFGKNEYIKCFEASS	377
Mb	VGGSLRHASNAAFVILCAAL.IGIM.SAQCFAFAFYCILDYLGDDGGSYVVGFGKNEYIKCFEASS	379
Of	VWGSIRHASNAAFVILCAAL.IGIM.SAQCFAFAFYCILDYLGDDGGSYVVGFGKNEYIKCFEASS	379
NL	KIGILCHAAANVAVFLICLCAAL.IGIM.SAQCFAFAFYCILDYLGDDGGSYVVGFGKNEYIKCFEASS	387
Ap	CLDFEAFQDWEAYRSEKPNFCVILGALVSGPDENDNYKDLFEYIYNEVTLIDYNAGFCSAVAGIHL	453
Bi	CPDFEAVCGWFEFDKDAE.CIILMGALVSGPDEADKFDHREDYVYTEVTLIDYNAGFTSALAGIHL	467
Ae	CPNKEFTECWCFCFSKNAPNFCIILMGALVSGPDEVDLEKDCFEYAYTEVTLIDYNAGFTSALAGIHL	463
Ca	CPDAEAVCDWTFNSELDNFHVITGALVSGPDCNDNYVDDFSDYVSEVATIDYNAGFCSAVAIVTL	446
Tc	CPNIEEFCQWQCFETWKGPNFCIILMGALVSGPDCNDHYEDVFEFELYNEVTLIDYNAGFCSTLAGIYI	444
Mb	CPFAEAVCDWSTYSSELDNFYVITGALVSGPDENDNYVDDFNDYVHNEVACIDYNAGFCSAVAIVTL	446
Of	CPDAEAVCDWSTYSSELDNFHVITGALVSGPDCNDNYVDDFNDYVQNVVACIDYNAGFCSAVSAIVTL	446
NL	CPNIEFVKCGWEAYNSTKDNFCVILGALVSGPDENDNYKDLFEYIYNEVTLIDYNAGFCSAVAGIHL	454
Ap	ELRASRHKAMMDANGLQLSSNATD	477
Bi	RVKSTI.....	473
Ae	QLKNVDTRPLKEN.....	476
Ca	GV.....	448
Tc	ENENKL.....	450
Mb	GY.....	448
Of	GY.....	448
NL	ELNNNS.....	461

Supplemental Figure S1. Protein alignment of GHF 9 enzymes from insects. Selected proteins accession numbers: Ae (*Acromyrmexechinator*, EGI63652), Ap (*Acyrtosiphonpisum*, XP_001944774), Bi (*Bombus impatiens*, XP_003494151), Ca (*Coptotermesacinaciformis*, AAK12339), Mb (*Macrotermesbarneyi*, AFD33365), Tc

(*Triboliumcastaneum*, EFA05721), Of (*Odontotermesformosanus*, ADB82658), NI (*Nilaparvatalugens*, NIEG1). The colors of the aligned sequences showed the degree of amino acid similarity, including identical (black), highly conserved (dark gray) and conserved residues (gray). The catalytic nucleophile, probable secondary nucleophile, and proton acceptor (Glu) of the conserved catalytic domain for GHF 9 are represented as solid diamond, open diamond, and solid circle, respectively. Two GHF 9 signature motifs are in bold below the alignment.



Supplemental Figure S2. Phylogenetic tree for amino acid sequences of NIEG1 and reported insect endogenous endo-β-1,4-glucanases. Selected proteins accession numbers: *Acromyrmexechinator* (XP_396791), *Acyrtosiphonpisum* (XP_001944774), *Apismellifera* (XP_396791), *Aprionagermari* (AAN78326), *Bombus impatiens* (XP_003494151), *Coptotermesacinaciformis* (AAK12339), *Coptotermesformosanus* (ACI75756), *Cryptocercusclevelandi* (AAF63720), *Nasutitermestakasagoensis* (BAA33708), *Neotermeskoshunensis* (BAD12005), *Odontotermesformosanus* (ADB82658), *Oncideresalbomarginatachamela* (ADI24131), *Panesthiacribrata* (AAF80584), *Psacotheahilaris* (BAB86867), *Reticulitemessperatus* (BAA31326), *Reticulitermesflavipes*(AAU20853), *Teleogryllusemma* (ABV32557), *Triboliumcastaneum* (EFA05721).



dsGFP



dsNIEG1

Supplemental Figure S3. The growth phenotype of BPH nymphs 6 days after they had been injected with either *GFP* (left) or *NIEG1* (right) dsRNA. Representative photographs are shown.

Supplemental Table S1. Primers and probes used for qRT-PCR and PCR.

Gene	Description	Forward primer (5'-...-3')	Reverse primer (5'-...-3')	Probe (5'-...-3')
<i>NIEG1</i>	qPCR	ATGATAGAGGGCTGAAGGGTG	TAGTAATGTCGTGGTGCTGGC	TGTCACCAGCGTCGTAGTAGCCTCC
<i>Actin</i>	qPCR	ATGAAACCGTCTACAACCTCG	GCATCCTGTCGGCAATAC	CGTCGACATCCGTAAGGACCT
<i>NIEG1</i>	Cloning	TGTAGGAATTGAGCAGCAAG	ATAAATTCTTTGGTGCATCC	
<i>NIEG1</i>	Protein expression	GGGGTACCAGCAAATACGACTACAGC	GCTCTAGAAATGAGTTGTCATTATTCA	
<i>NIEG1</i>	dsRNAsynthesis	GGATCCTAATACGACTCACTATAGGGCA	GGATCCTAATACGACTCACTATAGGACT	
		CGCTAAGCAACTCTT	CGTTCGGTCTGTCCT	
<i>GFP</i>	dsRNA synthesis	GGATCCTAATACGACTCACTATAGGAA	GGATCCTAATACGACTCACTATAGGCAG	
		GGGCGAGGAGCTGTTCACCG	CAGGACCATGTGATCGCGC	