Supporting Information

Systemically interfering with immune response by a fluorescent cationic

dendrimer delivered gene suppression

Dongxu Shen,^a Fan Zhou,^a Zejun Xu,^b Bicheng He,^a Miao Li,^a

Jie Shen,*a Meizhen Yin,*b and Chunju An*a

^a Department of Entomology, China Agricultural University, 100193 Beijing, China,

E-mail: shenjie@cau.edu.cn; anchunju@cau.edu.cn; anchunju@cau.edu.c

^b State Key Laboratory of Chemical Resource Engineering, Key Laboratory of Carbon Fiber and

Functional Polymers, Ministry of Education, Beijing University of Chemical Technology, 100029

Beijing, China, E-mail: <u>yinmz@mail.buct.edu.cn</u>

Gene	Forward primer	Reverse primer	
Amplification of the 821-bp fragment for serpin-3 labeling			
serpin-3	5'-TTGCCCACGAGTCCTACA-3'	5'-GCCATCGGCCAAGGTGGGGGTCG-3'	
Synthesis of dsRNA			
serpin-3	5'-	5'-	
	TAATACGACTCACTATAGGCTTCCAT	TAATACGACTCACTATAGGACACGCT	
	AACCCTCCTGC -3'	GTCCAAGTGCG -3'	
GFP	5'-	5'-	
	TAATACGACTCACTATAGGCACAAGT	TAATACGACTCACTATAGGGTTCACCT	
	TCAGCGTGTCCG -3'	TGATGCCGTTC -3'	
	Sequence for T7 promoter is underlined.		
<i>qRT-PCR analysis</i>			
serpin-3	5'-ATTGCAGCACAAATCGCCCC-3'	5'-GTGGGCAACTGCTGCAAACT-3'	
lectin	5'-GTCGTCGTACCTGGCCATCA-3'	5'-AGAAGTCGCCCTGGACATCG-3'	
PGRP	5'-TATGCGGGGGCATGCAGAACT-3'	5'-CCCATCCGCGACCTTCGTAT-3'	
βGRP	5'-GCCATGGCGCCTTTTGATGA-3'	5'-TCGTACCACGGCTTGGAGTC-3'	
rpL8	5'-AAGCGAGGAACATCAGCC-3'	5'-GGTCTTGCCACCACGAAT-3'	
cecropin	5'-CGCTTGTTTCATGGCGTTCG-3'	5'-ACGATGCCGTCTCGGATGTT-3'	
gloverin	5'-GGAGACCTCACTGCTGACCA-3'	5'-GTGCTCGTAGCCAGCTTTGC-3'	
moricin	5'-CCCTGCCCCTAAGGTTCCTG-3'	5'-CTGTAGACGTCGTGGGCTGT-3'	
Imd	5'-AGATCATGACAAACGAGGCAGT-3'	5'-TTGCTGCCAAACTGGACACC-3'	

1 88 ACTGGGCCGCTTGTGTGTAATCATGATTCAATAAACCAAATACCCTGTGCGGTGCTAACAGTGTTCAAAACTTCCAAAATAATAATAA 175 TTAAAAGGCCGGATCTCAATCGCCTTGAAGTTAATTTTTCTGTGTGTTTTCCTTATCAACATAATGACTCCAAGATTTTATTTCACA -19 MTPRF ΥF Т ${\tt CTTTTTGTAGTGGGCCCAGCACTATGTGTGTGGCCCAACAAATCGACCCGAACAACCGTGATCAACGTGTTTGGAACCCCCAGCAACTAC$ 262 -11 L F V V G P A L C V A O O I D P N T L I N V F G T P S N Y 349 GCTGCGTCTGTGAACCCCCATGACCCTGCGCGAGTGGAGTGCCGTTGGCCCAGGCCCTGCCGGAAACCCTATTAACGAAACCCTC 19 A A S V N P H D P A A S G V P L A Q A L P V K P I N E T L 436 ACTGATCCCGACTATTGGGATATAGATGAGCTGCCGGCAGCAGCGGTAGCTGACTATGATAAGTTTGACTGGAGTTTGACTAAGAGG 48 Т DPD Y W D I D E L P A A A V A D Y D K F D W S L TKR 523 77 L S A S S D T N F L L S P L G L K L A L A I L T E A A T G 610 106 R S E L Q S V L G F E M D R V A V R R K F A N I V N S L T 697 L O H K S P L Y V L D L G S K I Y V E N I A H P R O K F A 135 $\texttt{GCAG} \underline{\texttt{TTGCCCACGAGTCCTACA}} \texttt{AAACGGACCTGACTCCCATCGA} \underline{\texttt{CTTCCATAACCCTCCTGC}} \texttt{AGCCGCCAAAGCCATCAACGATTGG}$ 784 164 V Α Η Е S Y KTDLTPID F Η N P Ρ АААКА I N D W 871 ${\tt GTTGCCAACCTTACCCAGGGAAGGATCACTGACTTAGTACATCAAGATGACCTCGAAAACGTGGTGGTGGTGATGATCCTCAACACACTA$ 193 V A N L T Q G R I T D L V H Q D D L E N V V VMILNTL 958 TACTTCAAGGGCAGCTGGCGCCCAGCTCGCGCCCAACGCTACCAAGCAGGGTCAATTCTACGTCACCCCGAAAATAGCTAAACCA 222 Y F K G S W R H O F A P N A T K O G O F Y V T P K I A K P 1045 GTGTATTTCATGAATGTGAAGGACAAGTTCTACTATGCTGAGTCTGCCAAATTCGACGCTAAGATCCTCAGGATGCCATACATGGGC 251 YAESAKFDAKILRM V Y F M N V K D K F Y Ρ Y M G 1132 TACAAATTTGCAATGTACGTAGTAGTTCCCAACTCATTGACTGGTCTGAACCGAGTATTGGATGGTCTGACGGAGCTCCGCCCCGAA 280 K F A M Y V V P N S L T G L N R V L D G L T E L R P E ATGGATTTGTTACAAGAGCGCCTCGTCGACGTCACTTTGCCCAGATTCCAGTTCGAATTCTCCT<u>CGCACTTGGACAGCGTGT</u>TGAGA 1219 M D L L Q E R F V D V T L P R F Q F E F S S H L 309 S V L R D 1306 GATATGGGTGTCAGACAAGCCTTCGAGGACACTGCGTCGTTCCCTGGCATCGCCAGGGGACAGTCCCTGCAGCAGCGCCTCAGGGTC 338 D M G V R Q A F E D T A S F P G I A R G Q S L Q Q R L R V ${\tt TCCAAGGTTCTGCAGCGGTCGGGCATCGAGGTCAACGAGCTCGGAAGTGTGGCCTACTCAGCTACTGAAATATCTCTAGTCAACAAA$ 1393 S K V L O R S G I E V N E L G S V A Y S A T E I S L V N 367 Κ TTCGGCGAGGATGACGATACCGCTGTGGAGGTGATCGCCAACAAGCCCTTCTTCTTCCTGATCCAAGATGAGACGACCAGGCAACTG 1480 F G E D D D T A V E V I A N K P F F F L I Q D E T T R Q L 396 1567 LFTGRVADPTLADGTFKHS* 425 $1654 \quad {\tt TAGTAATCGCCCGACTGAAACTTCCTTGATTCTTAAGTTAAAATTTCCCACAGGTCTTGCTTTGGGTAATTTTGTTTTGCAACTGAACT$ 1741 TTGATAATGCCCTGAAGTGTATTACTCGTATAATTGTTTTTCTAATAGCTTATGATAATGTGGTCAGCTTTTAAACCCCATCTTACGT 1828 GGCCAGAAGTGCTAGAATATTGTAGATAGGATACAGCTATCGTTTAAGTTTTGGAAAGGTGATTACTGAGAGGGGCATAATATTGTTT 2002 GTACATGATGGCTCTCTTTTGATTCGATCTGTGATCTGTTTAACATTTATACGAAGTAAAAATATATCAAAAACCAAGGTTATTGAAG

Figure S1. Nucleotide and amino acid sequence of *O. furnacalis* serpin-3. The deduced amino acid sequence is shown below the nucleotide sequence of serpin-3 (GenBankTM accession number KF501490). The one-letter code for each amino acid is aligned with the second nucleotide of the corresponding codon. The stop codon is marked with *asterisk* (*). The primers for amplifying *serpin-3* DNA and dsRNA fragment are underlined and double-underlined, respectively.



Figure S2. Absorption and emission spectra of G2 in water (concentration: 2×10^{-6} M, $\lambda_{ex} = 545$

nm).



Figure S3. Fluorescence images of G2/DNA complexes (N/P= 2:1) after 8 h incubation with *O. furnacalis* midgut (A1-A5), fat body (B1-B5), and hemocyte (C1-C5). A1, B1 and C1: separated channels for tissues or cells only. A2, B2, and C2: separated channels for G2 (red). A3, B3, and C3: separated channels for DNA labeled by CXR Reference Dye (blue). A4, B4, and C4: merged images from the second and third one on the left. A5, B5, and C5: merged images from the three ones on the left. Gr: granulocyte



Figure S4. Agarose gel electrophoresis of G2/DNA complexes at various N/P ratios.

Table S2. Particle sizes of G2 before and after it forms a complex with DNA at N/P = 8:1.

	Average size(nm)
G2	1.8 ± 0.3
DNA	1318.3±3.2
G2/DNA	136.5 ± 3.4

Table S3. The zeta-potentials of G2 before and after it form complexes with DNA at N/P = 8:1.

	Zeta potential
	(mV)
G2	13.3 ± 0.8
DNA	-24.2 ± 0.5
G2/DNA	11.6 ± 1.2