**Supplementary Material**

# Silencing of multiple Target Genes via ingestion of dsRNA and PMRi affects development and survival in *Helicoverpa armigera*

**Muhammad Nauman Sharif[1], Muhammad Shahzad Iqbal[2], Rukkaya Alam[3], Mudassar Fareed Awan[4], Rao Muhammad Tariq[1], Qurban Ali[5*]and Idrees Ahmad Nasir[1]**

1. Center of Excellence in Molecular Biology, University of the Punjab, Lahore, Pakistan.
2. Department of Biotechnology, Faculty of Life Sciences, University of Central Punjab, Lahore.
3. Department of Zoology, University of the Punjab, Lahore, Pakistan.
4. University of Management and Technology, Sialkot, Pakistan
5. Institute of Molecular Biology Biotechnology, The University of Lahore, Lahore, Pakistan.

Corresponding Author Email address: nauman.cemb@gmail.com, saim169@gmail.com

**Primers used**

| Primer Name | Sequence 5′ to 3′ | Purpose |
|---|---|---|
| AchE-F | TTCAAACGAGGCATCTTGC | Amplification |
| AchE-R | TTGCCAGTCAAAGCGAATC | Amplification |
| EcR-F | TCCAGCTTCAAGTGTGAACG | Amplification |
| EcR-R | CATATCCGAGTCCTCGTCGT | Amplification |
| vAA-F | TATCCTGGGCTCCATCTTTG | Amplification |
| vAA-R | GACAGGCATGTTGGATGTGT | Amplification |
| AT-F | TCTCAATGCACCTGGCAGTA | Amplification |
| AT-R | AACGAGATGCTGAACAACCC | Amplification |
| AS-F | GAAATCAAGCGCGTACAACG | Amplification |
| AS-R | CCAGTGCTACTTCAACCCCA | Amplification |
| L4AT-F | GGCAAGCTTGCGTGACAGACCACACACTA | L4440 cloning |
| L4AT-R | GGCGAGCTCGGGTTGTTCAGCATCTCGTTC | L4440 cloning |
| L4AS-F | GGCAAGCTTTCACAGTAAACGCTGCACCT | L4440 cloning |
| L4AS-R | GGCGAGCTCTGGGGTTGAAGTAGCACTGG | L4440 cloning |
| L4AchE-F | GGCAAGCTTCCGGACACAATGATGAAAGA | L4440 cloning |
| L4AchE-R | GGCGAGCTCCCCTGGTGTGGTATTGAAGG | L4440 cloning |
| L4EcR-F | GGCAAGCTTGGCAGCACGAAGAGCTATGT | L4440 cloning |
| L4EcR-R | GGCGAGCTCCCTTGCGATCAACGACTTCT | L4440 cloning |
| L4vAA-F | GGCAAGCTTTCAACGAGCTCACACAGTCC | L4440 cloning |
| L4vAA-R | GGCGAGCTCCACAATGACATCGGAGTTGG | L4440 cloning |
| L4egfp-F | GGCAAGCTTCACATGAAGCAGCACGACTT | L4440 cloning |
| L4egfp-R | GGCGAGCTCTGCTCAGGTAGTGGTTGTCG | L4440 cloning |
| TAchE-F | GGCAAGCTTCCGGACACAATGATGAAAGA | TRV cloning |
| TAchE-R | GGCGGATCCCCCTGGTGTGGTATTGAAGG | TRV cloning |
| TEcR-F | GGCAAGCTTGGCAGCACGAAGAGCTATGT | TRV cloning |
| TEcR-R | GGCGGATCCCCTTGCGATCAACGACTTCT | TRV cloning |
| TvAA-F | GGCAAGCTTTCAACGAGCTCACACAGTCC | TRV cloning |

| | | |
|---|---|---|
| TvAA-R | GGCGGATCCCACAATGACATCGGAGTTGG | TRV cloning |
| PAchE-F | CGACTGAATCCGGTGAGAAT | Plant detection |
| PAchE-R | GGTTGCATTCGATTCCTGTT | Plant detection |
| PEcR-F | AAATGCTTGATGGTCGGAAG | Plant detection |
| PEcR-R | TCAGGTGCGACAATCTATGG | Plant detection |
| PvAA-F | GGTTGCATTCGATTCCTGTT | Plant detection |
| PvAA-R | ATTCAACGGGAAACGTCTTG | Plant detection |
| qAchE-F | ACGAGATTCGCTTTGACTGG | qRT-PCR |
| qAchE-R | ATCATTCCAGAAAGCGCAAG | qRT-PCR |
| qEcR-F | GGTGCTCGACTCACTCTTCC | qRT-PCR |
| qEcR-R | CTGACATCGGAGGTGCAGTA | qRT-PCR |
| qvAA-F | CTGACGTAGTGCTGGAGACG | qRT-PCR |
| qvAA-R | AGGGAAGAGTGAGTCGAGCA | qRT-PCR |
| qegfp-F | AGAACGGCATCAAGGTGAAC | qRT-PCR |
| qegfp-R | TGCTCAGGTAGTGGTTGTCG | qRT-PCR |
| q18s-F | GCCTCTTTGTCCAGATCAGC | qRT-PCR |
| q18s-R | ACTTCTTGGCTTTGGCAGAA | qRT-PCR |
| qβAct-F | TGCGTGACATCAAGGAGAAG | qRT-PCR |
| qβAct-R | TACCGATGGTGATGACCTGA | qRT-PCR |

**Sequences of cloned gene fragments.**

**v-ATPase-A**

TCAACGAGCTCACACAGTCCATCTACATCCCCAAGGGTGTAAACGTACCTTCTCTGGCTAG
GGGTGTCAGCTGGGAATTCGTTCCCGTGAGTGTTAAGACGGGCTCCCACATCACTGGCGGA
GACCTGTACGGTCTGGTGCACGAGAACACGCTGGTGAAGCACCGCGTGTTGATCCCGCCCA
AGGCCAAGGGTACCGTCACATACATCGCGCCCGCTGGCAACTACAAAGTCACTGACGTAGT
GCTGGAGACGGAGTTCGACGGCGAGAAGGAGAAGTACACGGTGTTGCAGGTGTGGCCGGT
GCGTCAGCCGCGACCCGTCACCGAGAAGCTCCCCGCCAACCATCCGCTGCTCACCGGGCAG
AGGGTGCTCGACTCACTCTTCCCTTGCGTCCAGGGAGGTACCACTGCCATTCCCGGAGCTTT
CGGTTGCGGCAAGACTGTCATCTCGCAGGCGCTGTCCAAGTACTCCAACTCCGGTGTCATT
GTG

**Ecdyson Receptor**

GGCAGCACGAAGAGCTGTGTCTAGTCTGCGCCGACAGAGCCTCCGGATATCACTACAAAGC
ACTCACGTGTGAACGGTGTAAAGGTTTGTTCAGGCGGAGTGTAACCCAAAGTGCAGTGTAC
GTGTGCAAATTCGGCAGTGCTTGCGGAGTGGGTGTGTGTGTGCGGAGAAAGTGTCAGGAGT
GTAGGTTGAAGAAAGGTCTCGCGGTAGGCGTGAGGCCGGAGTGCGTGGTGCCGGAGAACC
AGTGTGCAGTGAAACGGAAAGAGGAAAAGGCGCAGAGGGAAAAGGACCAATTGCCCGTC

AGTACGACGACAGTAGACGATCACGTGCCTCCCATCGTGCAGTGTGATCCTCCGGGCCCAG
AGGCCGCTAGAATTCACGAGGTGGTGCCGCGGTTCCTCAGTGAGAAGCTCGTGGAACAGA
ACAGATTGAAGAAGGTGGCCCCCCTCACTGCCAATCAGAAGTCGTTGATCGCAAGG

**Acetylcholine Esterase**

CCGGACACAGTGGTGAAAGAAGGCAATTTCCATAATACCGAGGTGCTTCTTGGAAGTAATC
AAGACGAAGGAACCTATCTCTTACTATACGATTCCCTTGACTACTTCGAGAAAGACGGCAC
CAGCTCCTTGCAGCGGAAGCAAATTCTAGAAATTGTCGACACCATATTCAAGGATTTCTCC
AAAATAAAGAGCCAAGCTATCGTATTCCAATATACGGACTGGCAGGAAAATACCGGTGGC
TATCTGAACCAGGAAGTGATAGCTGACGTGGTGGGCGACTGTGTCTACGTGTGCCCTACTA
ACTACTTCGCAGAGGTGCTGGCCGATTCAGGAGTTGGTGTCCACTACTATTACTTCACACAT
CGCACCAGCACAAGTCTCTTGGGTGAGTGGATTGGCGTGGTGCAAGGCGGTGAGGTGGAGT
ACGTCTTCCGGCACCCGCTGAACGTGTTCCTTCAATACCACACCAGCG

**egfp**

CCACGTGAAGCAGCACGACTTCTTCAAGTCCGCCGTGCCCGAAGGCTACGTCCAGGAGCGC
ACCATCTTCTTCAAGGACGACGGCAACTACAAGACCCGCGCCGAGGTGAAGTTCGAGGGC
GACACCCTGGTGAACCGCATCGAGCTGAAGGGCATCGACTTCAAGGAGGACGGCAACATC
CTGGGGCACAAGCTGGAGTACAACTACAACAGCCACAACGTCTATATCGTGGCCGACAAG
CAGAAGAACGGCATCAAGGTGAACTTCAAGATCCGCCACAACATCGAGGACGGCAGCGTG
CAGCTCGCCGACCACTACCAGCAGAACACCCCCATCGGCGACGGCCCCGTGCTGCTGCCCG
ACAACCACTACCTGAGCA