

Rht1-A ATGAAGCGGAGTACCAGGACGCCGGCGGAGCGGTGGCGGGG--CGGCATGGGCTCG 57
 Rht1-B ATGAAGCGGAGTACCAGGACGCCGGAGGAGCGCGGTGGCGGGGAGGCATGGGCTCG 60
 Rht1-D ATGAAGCGGAGTACCAGGACGCCGGAGGAGCGCGCGGTGGCGGCATGGGCTCG 60

 Rht1-A TCTGAGGACAAGATGATGGTGTCTCG-----CGGCGCGGGGGAGGGGAGGAGGTGGAC 111
 Rht1-B TCCGAGGACAAGATGATGGTGTCTCGGGTTCGGCGCGCGCGGGGGAGGGGAGGAGGTGGAC 120
 Rht1-D TCCGAGGACAAGATGATGGTGTCTCG-----CGGCGCGGGGGAGGGGAGGAGGTGGAC 114
 ** *****
 Rht1-A GAGCTGCTGGCGGCGCTCGGGTACAAGGTGCGGCTCCGACATGGCGGACGTGGCGCAG 171
 Rht1-B GAGCTGCTGGCGGCGCTCGGGTACAAGGTGCGGCTCCGACATGGCGGACGTGGCGCAG 180
 Rht1-D GAGCTGCTGGCGGCGCTCGGGTACAAGGTGCGGCTCCGACATGGCGGACGTGGCGCAG 174

 Rht1-A AAGCTGGAGCAGCTGGAGATGGCCATGGGGATGGGCGGCGTGGGCGCCGGCGCGCCCC 231
 Rht1-B AAGCTGGAGCAGCTGGAGATGGCCATGGGGATGGGCGGCGTGGGCGCCGGCGCGCCCC 240
 Rht1-D AAGCTGGAGCAGCTCGAGATGGCCATGGGGATGGGCGGCGTGGGCGCCGGCGCGCCCC 234

 Rht1-A GACGACAGCTTCGCCACCCACCTCGCCACGACACCGTGCACCTACAACCCACCGACCTC 291
 Rht1-B GACGACAGCTTCGCCACCCACCTCGCCACGACACCGTGCACCTACAACCCACCGACCTC 300
 Rht1-D GACGACAGCTTCGCCACCCACCTCGCCACGACACCGTGCACCTACAACCCACCGACCTG 294

 Rht1-A TCCTCCTGGGTCGAGAGCATGCTGTCTGGAGCTCAACGCGCCGCGCGCCCCCTCCCGCCC 351
 Rht1-B TCCTCCTGGGTCGAGAGCATGCTGTCTGGAGCTCAACGCGCCGCGCGCCCCCTCCCGCCC 360
 Rht1-D TCCTCCTGGGTCGAGAGCATGCTGTCTGGAGCTCAACGCGCCGCGCGCCCCCTCCCGCCC 354
 ** *****
 Rht1-A GCCCGCAGCAGCTCAACGCCTCCACCTCCTCCACCGTCACGGC--GGTGGGTACTTC 408
 Rht1-B GCCCGCA--GCTCAACGCCTCCACCTCCTCCACCGTCACGGC--GGTGGGTACTTC 414
 Rht1-D GCCCGCA--GCTCAACGCCTCCACCTCCTCCACCGTCACGGCAGCGCGGGTACTTC 411

 Rht1-A GATCTCCCGCCCTCGGTGACTCCTCTCAGCACCTACGCCCTGCGGCGGATCCCTCC 468
 Rht1-B GATCTCCCGCCCTCGGTGACTCCTCTCAGCACCTACGCCCTGCGGCGGATCCCTCC 474
 Rht1-D GATCTCCCGCCCTCGGTGACTCCTCCAGCAGATCTACGCGTTCGCGCGGATCCCTCC 471

 Rht1-A CCGGCGGCGCGCGTCCGGCCGCGACCTGTCCGCGACTCCGT--GCGGGACCCCAAG 525
 Rht1-B CCGGCGGCGCGCGTCCGGCCGCGACCTGTCCGCGACTCCGTGCGGCGGATCCCAAG 528
 Rht1-D CCGGCGGCGCGCGTCCGGCCGCGACCTGTCCGCGACTCCGT--GCGGGATCCCAAG 528

 Rht1-A CGGATGCGCACTGGCGGGAGCAGCACCTCGTCGTCGTCATCTCTCTCTCTCTCGG- 584
 Rht1-B CGGATGCGCACTGGCGGGAGCAGCACCTCGTCGTCATCTCTCTCTCTCTCTCGG 585
 Rht1-D CGGATGCGCACTGGCGGGAGCAGCACCTCGTCGTCATCTCTCTCTCTCTCTCTCGG- 587

 Rht1-A --TGGGGCGCCAGGAGCTCTGTGGTGGAGGCTGCTCCGCCGTCGCGGCCGGGGCCAAC 642
 Rht1-B GGTGGCGGCGCCAGGAGCTCTGTGGTGGAGGCTGCCCCGCCGTTGGCGCCGGCGCGGT 645
 Rht1-D --TGGGGCGCCAGGAGCTCTGTGGTGGAGGCTGCCCCGCCGTCGCGGCCGGGGCCAAC 645
 *** *****
 Rht1-A GCG--CCGCGCTGCGCGTCTGTGGTGGTTCGACACGAGGAGCGGGATTTCGGCTGGTG 699
 Rht1-B GCG--CCGCGCTGCGCGTCTGTGGTGGTTCGACACGAGGAGCGGGATTTCGGCTGGTG 702
 Rht1-D GCGACGCCCGCTGCGCGTCTGTGGTGGTTCGACACGAGGAGCGGGATTTCGGCTGGTG 705
 *** *****
 Rht1-A CACGCGTGTGCGCTGCGCGGAGGCGGTGAGCAGGAGAATCTCTTGCCGCGGAGGCG 759
 Rht1-B CACGCGTGTGCGCTGCGCGGAGGCGGTGAGCAGGAGAATCTCTTGCCGCGGAGGCG 762
 Rht1-D CACGCGTGTGCGCTGCGCGGAGGCGGTGAGCAGGAGAATCTCTTGCCGCGGAGGCG 765

 Rht1-A CTGGTGAAGCAGATACCTTGTGGCCGCGTCCAGGGCGCGCGATGCGCAAGGTTCGCC 819
 Rht1-B CTGGTGAAGCAGATACCTTGTGGCCGCGTCCAGGGCGCGCGATGCGCAAGGTTCGCC 822
 Rht1-D CTGGTGAAGCAGATACCTTGTGGCCGCGTCCAGGGCGCGCGATGCGCAAGGTTCGCC 825

 Rht1-A GCCTACTTCGGCGAGGCCCTCGCCCGCGCGTCTTCGCTTCGCGCCGAGCCGACAGC 879
 Rht1-B GCCTACTTCGGCGAGGCCCTCGCCCGCGCGTCTTCGCTTCGCGCCGAGCCGACAGC 882
 Rht1-D GCCTACTTCGGCGAGGCCCTCGCCCGCGCGTCTTCGCTTCGCGCCGAGCCGACAGC 885

 Rht1-A TCCCTCCTCGACGCGCGCTTCGCCGACCTCCTCCACGCGCACTTCTACGAGTCTGCCCC 939
 Rht1-B TCCCTCCTCGACGCGCGCTTCGCCGACCTCCTCCACGCGCACTTCTACGAGTCTGCCCC 942
 Rht1-D TCCCTCCTCGACGCGCGCTTCGCCGACCTCCTCCACGCGCACTTCTACGAGTCTGCCCC 945

 Rht1-A TACCTCAAGTTTCGCGCACTTCACCGCAACCAGGCCATCCTGGAGGCGTTCGCGCGGTGC 999
 Rht1-B TACCTCAAGTTTCGCGCACTTCACCGCAACCAGGCCATCCTGGAGGCGTTCGCGCGGTGC 1002
 Rht1-D TACCTCAAGTTTCGCGCACTTCACCGCAACCAGGCCATCCTGGAGGCGTTCGCGCGGTGC 1005

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Rht1-A CGCCGCGTGCACGTCGTGACTTCGGCATCAAGCAGGGGATGCAGTGGCCCGCCTCTC 1059
Rht1-B CGCCGCGTGCACGTCGTGACTTCGGCATCAAGCAGGGGATGCAGTGGCCCGCCTCTC 1062
Rht1-D CGCCGCGTGCACGTCGTGACTTCGGCATCAAGCAGGGGATGCAGTGGCCCGCACTCTC 1065
*****

Rht1-A CAGGCCCTGCGCTCCGTCCCGCGGCCCTCCCTCGTTCGCGCTCACCGGCGTCGGCCCC 1119
Rht1-B CAGGCCCTGCGCTCCGTCCCGCGGCCCTCCCTCGTTCGCGCTCACCGGCGTCGGCCCC 1122
Rht1-D CAGGCCCTGCGCTCCGTCCCGCGGCCCTCCCTCGTTCGCGCTCACCGGCGTCGGCCCC 1125
*****

Rht1-A CCGCAGCCGACGAGACCGACGCCCTTGCAGCAGGTGGGTGGAAGCTCGCCAGTTCGCG 1179
Rht1-B CCGCAGCCGACGAGACCGACGCCCTTGCAGCAGGTGGGTGGAAGCTCGCCAGTTCGCG 1182
Rht1-D CCGCAGCCGACGAGACCGACGCCCTTGCAGCAGGTGGGTGGAAGCTCGCCAGTTCGCG 1185
*****

Rht1-A CACACCATCCGCGTCGACTTCCAGTACCGCGGCCCTCGTCGCCGCCACGCTCGCGGACCTG 1239
Rht1-B CACACCATCCGCGTCGACTTCCAGTACCGCGGCCCTCGTCGCCGCCACGCTCGCGGACCTG 1242
Rht1-D CACACCATCCGCGTCGACTTCCAGTACCGCGGCCCTCGTCGCCGCCACGCTCGCGGACCTG 1245
*****

Rht1-A GAGCCATTCATGCTGCAGCCGAGGGCGAGGAGGCCGAACGAGGAGCCCGAGGTAATC 1299
Rht1-B GAGCCATTCATGCTGCAGCCGAGGGCGAGGAGGCCGAACGAGGAGCCCGAGGTAATC 1302
Rht1-D GAGCCATTCATGCTGCAGCCGAGGGCGAGGAGGCCGAACGAGGAGCCCGAGGTAATC 1305
*****

Rht1-A GCCGTCAACTCGGTCTCGAGATGCACCGGCTGCTCGCGCAGCCCGCGGCCCTGGAGAAG 1359
Rht1-B GCCGTCAACTCGGTCTCGAGATGCACCGGCTGCTCGCGCAGCCCGCGGCCCTGGAGAAG 1362
Rht1-D GCCGTCAACTCGGTCTCGAGATGCACCGGCTGCTCGCGCAGCCCGCGGCCCTGGAGAAG 1365
*****

Rht1-A GTCTGGGACCGTGCAGCGCGTGCAGGCCAGGATCGTCACCGTGGTGGAGCAGGAGGCC 1419
Rht1-B GTCTGGGACCGTGCAGCGCGTGCAGGCCAGGATCGTCACCGTGGTGGAGCAGGAGGCC 1422
Rht1-D GTCTGGGACCGTGCAGCGCGTGCAGGCCAGGATCGTCACCGTGGTGGAGCAGGAGGCC 1425
*****

Rht1-A AACCACAATCCGGCACATTCCTGGACCGCTTACCGAGTCTCTGCACTACTACTCCACC 1479
Rht1-B AACCACAATCCGGCACATTCCTGGACCGCTTACCGAGTCTCTGCACTACTACTCCACC 1482
Rht1-D AATCACAATCCGGCACATTCCTGGACCGCTTACCGAGTCTCTGCACTACTACTCCACC 1485
*****

Rht1-A ATGTTTCGATTCTCTGAGGGCGGCAGCTCCGGCGGC--CCATCCGAAGTCTCATCGGG 1536
Rht1-B ATGTTTCGATTCTCTGAGGGCGGCAGCTCCGGCGGC--CCATCCGAAGTCTCATCTGG 1539
Rht1-D ATGTTTCGATTCTCTGAGGGCGGCAGCTCCGGCGGCAGCTCCGAAGTCTCATCGGG 1545
*****

Rht1-A GCTGCCGCTGCTCCTGCCGCCGCCGGCACGGACAGGTGATGTCGAGGTGTACCTCGGC 1596
Rht1-B GCGGCTGCTGCTCCTGCCGCCGCCGGCACGGACAGGTGATGTCGAGGTGTACCTCGGC 1599
Rht1-D GCTGCTGCTGCTCCTGCCGCCGCCGGCACGGACAGGTGATGTCGAGGTGTACCTCGGC 1605
*****

Rht1-A CGGCAGATCTGCAACGTGGTGGCCTGCGAGGGGGCGGAGCGCACAGAGCGCCACGAGACG 1656
Rht1-B CGGCAGATCTGCAACGTGGTGGCCTGCGAGGGGGCGGAGCGCACAGAGCGCCACGAGACG 1659
Rht1-D CGGCAGATCTGCAACGTGGTGGCCTGCGAGGGGGCGGAGCGCACAGAGCGCCACGAGACG 1665
*****

Rht1-A CTGGGGCAGTGGCGGAACCGGCTGGGCAACGCCGGGTTCGAGACCGTGACCTGGGCTCC 1716
Rht1-B CTGGGGCAGTGGCGGAACCGCCTCGGCAACGCCGGGTTCGAGACCGTGACCTGGGCTCC 1719
Rht1-D CTGGGCCAGTGGCGGAACCGGCTGGGCAACGCCGGGTTCGAGACCGTGACCTGGGCTCC 1725
*****

Rht1-A AATGCCCTACAAGCAGGCGAGCACGCTGCTGGCCCTATTCGCCGGCGGCGACGGGTACAAG 1776
Rht1-B AATGCCCTACAAGCAGGCGAGCACGCTGCTGGCGCTCTTCGACGGCGGCGACGGGTACAAG 1779
Rht1-D AATGCCCTACAAGCAGGCGAGCACGCTGCTGGCGCTCTTCGCCGGCGGCGACGGGTACAAG 1785
*****

Rht1-A GTGGAGGAGAAGGAGGGCTGCCTGACTCTGGGTGGCACACGCGCCCGTGATCGCCACC 1836
Rht1-B GTGGAGGAGAAGGAGGGCTGCCTGACTCTGGGTGGCACACGCGCCCGTGATCGCCACC 1839
Rht1-D GTGGAGGAGAAGGAGGGCTGCCTGACTCTGGGTGGCACACGCGCCCGTGATCGCCACC 1845
*****

Rht1-A TCGCATGGCGCCTGGCCGCGCCGTGA 1863
Rht1-B TCGCATGGCGCCTGGCCGCGCCGTGA 1866
Rht1-D TCGCATGGCGCCTGGCCGCGCCGTGA 1872
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Additional Data File 3 Sequence alignment of three copies of *Rht1* gene (A, B, D copies). The regions of the gene covered by the sequenced reads from mutant and wild plants are shown by colored lines. Homoeologous SNPs are depicted in yellow whereas position of EMS induced SNPs are highlighted in cyan. Underlined SNP indicated detection both by redefined as well as first criteria whereas non-underlined SNP indicated detection with redefined criteria only.