GENETICS

Supporting Information

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Miniature Inverted-Repeat Transposable Elements of *Stowaway* Are Active in Potato

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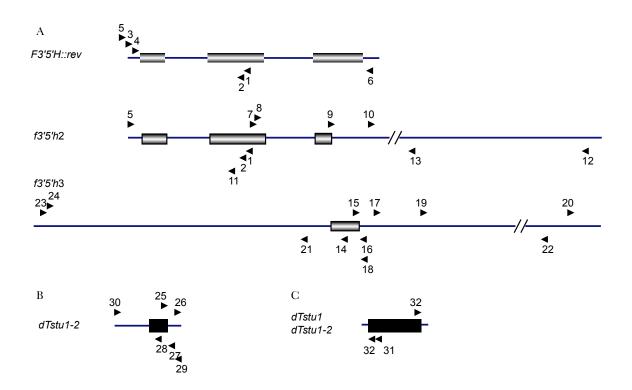


FIGURE S1.—Approximate positions of primers used in this study. Arrow heads show primers and their directions used for the analysis of (A) F3'5'H (pseudo) genes as described in supplementary methods, (B) the insertion locus of dTstu1-2 and (C) MITE display. Coding regions are marked by shaded boxes and the *Stowaway* MITE is indicated by a black bar.

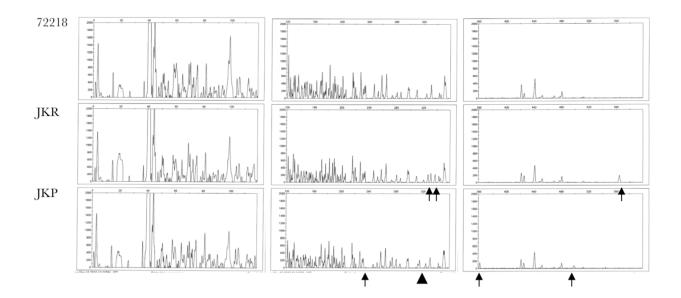


FIGURE S2.—MITE display using primers designed from sequences of *dTstu1* and *dTstu1-2*. Mse+T was used as a selective primer. The left panels show peaks from 0 base to 120 bases, the middle from 120 to 360 bases and the right from 360 to 600 bases. Differences of 'JKR' and 'JKP' as compared with '72218' are indicated by arrows. An arrowhead shows a new peak of 315 bases in size.

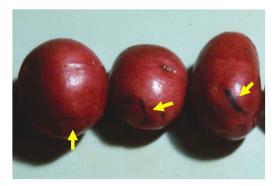


FIGURE S3.—Tuber appearance of one of the plants regenerated from protoplasts of '72218'. Arrows indicate purple variegation (which appear as a dark line in the picture) present against the red background.

FILE S1

Supporting Methods

Isolation and sequences determination of the genomic DNA for F3'5'H genes: For the isolation of F3'5'H pseudo-gene, J3'5'h2, inverse PCR was carried out with primers No. 1 and 7 for the 1st, and No. 2 and 8 for the 2nd amplification using as a template self-ligated genomic DNA from 'JKP' which had been digested by *Mbol*. Primers No. 7 and 8 were based on the highly conserved region among P450 or F3'5'H genes. The product was cloned and its sequence enabled the design of primers No. 9, 10 and 11, which were used in inverse PCR (with primers No. 2 and 9 for the 1st and No. 10 and 11 for the 2nd amplification) on self-ligated *Xbal*-digested genomic DNA of 'JKP'. The sequence of the resulting 5 kb product provided information to design primers No. 12 and 13 that were used in PCR reactions on genomic DNA from 'JKP' and '72218' to compare their J3'5'h2 alleles (primers No. 5 and 12 for the 1st and No. 5 and 13 for the 2nd amplification). The third copy of F3'5'H pseudo-gene, J3'5'h3, was isolated after a series of inverse PCR reactions that yielded sequence to design primers that were used in subsequent rounds. Inverse PCRs were done on *Hin*cII-digested genomic DNA followed by self-ligation with primers No. 14 and 15 and then self-ligated *Eco*RI-digested DNA with a primer set of No. 16 and 17 for the 1st followed by a set of No. 17 and 18 for the 2nd amplification. The penultimate round of inverse PCR was performed on *Hin*dIII-digested and self-ligated DNA with primers No. 18 and 19 for the 1st, and No. 20 and 21 for the 2nd amplifications. Finally, primers could be designed (No. 22 and 23 for the 1st and No. 18 and 24 for the 2nd amplification) to amplify J3'5'h3 from both 'JKP' and '72218' genomic DNAs as templates (see Figure S1). The extension time in all PCRs was 5 min.

TABLE S1

Primer sequences used in this study

| Duin | |
|--------|---|
| Primer | Sequence |
| 1 | |
| 1 | 5'-AACATTTTTGTCAATAAAKCATCAAA-3' |
| 2 | 5'-CCTTGTAAATCCATCCAAGCTA-3' |
| 3 | 5'-CCGAATTCAAGCTTTATATTATTATCTTCGATTTT-3' |
| 4 | 5'-GGCATTACGTATTAGTGAGTTG-3' |
| 5 | 5'-CCTTCTACTTCATTCTCACTCT-3' |
| 6 | 5'-AGCAAATATGTTGCACTATAAATG-3' |
| 7 | 5'-CCTGATTTTCTTGATKTTRTTATGG-3' |
| 8 | 5'-GGGATAATTCTGAAGGAGAAAG-3' |
| 9 | 5'-TATTCCAAGTTGTTGACACCCA-3' |
| 10 | 5'-ACTGAAGTAGCCATCCAAAGAC-3' |
| 11 | 5'-TCAACGAACACTCTCTTACTTAA-3' |
| 12 | 5'-GCTCACTACACAATGCACATG-3' |
| 13 | 5'-TCATGAAATGCATCGACAATTTAT-3' |
| 14 | 5'-GCTAATCCAAAAGATTCCTCCA-3' |
| 15 | 5'-TGCAAAAAGCTGTCCCTCTTG-3' |
| 16 | 5'-TTACGTTACGGTCTTCAACAG-3' |
| 17 | 5'-AGAGGAGGATAACAAACTTGTAT-3' |
| 18 | 5'-AACAGATACGTTGCACTATAACT-3' |
| 19 | 5'-CCTAGTCCCCATTTCACTACA-3' |
| 20 | 5'-GAACATGAGTTTACGTGAACCC-3' |
| 21 | 5'-AGGCATCCTTCGAAATCCACA-3' |
| 22 | 5'-TTGTAAAGTGCACCCATCATCT-3' |
| 23 | 5'-CATTCGTCCTAACGATGGACA-3' |
| 24 | 5'-ATTCAGATCCTCCCGATGAATT-3' |
| 25 | 5'-ATTCATTTTGGACCACAAGTTTTA-3' |
| 26 | 5'-TGTTTTTTGCAGTTATCTTATTTCA-3' |
| 27 | 5'-CAAGGGGAGACATTTAGG-3' |
| 28 | 5'-AGACATTTCATAGGCAAATTGTTA-3' |
| 29 | 5'-AGCTGAAATATGAGATTGAAATTAG-3' |
| 30 | 5'-ATTTTGCTATATCCACAATGACTT-3' |
| 31 | 5'-CATTCTTTTTGGGACTGACTA-3' |
| 32 | 5'-ATAAAWTGGGACRGAGGGAGTA-3' |
| Mse+0 | 5'-GACGATGAGTCCTGAGTAA-3' |
| Mse+N | |
| | |