

Supplementary material to:

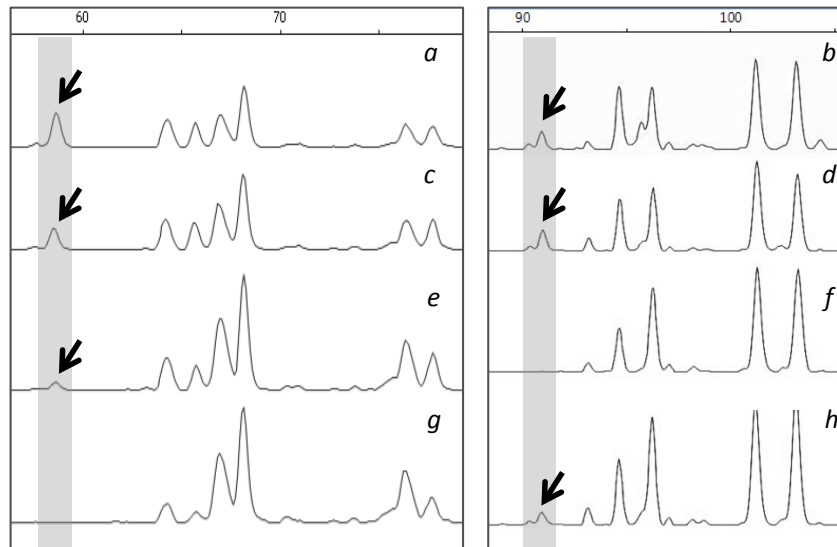
Stochastic changes affect *Solanum* wild species following autopolyploidization

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Case	5' C C G G 3'				Banding pattern	
	3' G G C C 5'				<i>HpaII</i>	<i>MspI</i>
1. Null methylation	○	○	□	□	+	+
2. Full methylation	●	●	□	□	-	-
3. Full CHG methylation	●	○	□	□	-	-
4. Hemi CHG methylation	●	○	□	□	+	-
5. Hemi CG methylation	○	●	□	□	-	+
6. Full CG methylation	○	●	□	□	-	+

○ = unmethylated Cytosine ● = methylated Cytosine □ = Guanine
+ = presence of the band - = absence of the band

Supplementary Fig 1. Possible methylation states at 5'CCGG sites, and the corresponding banding patterns produced by the isoschizomers *HpaII* and *MspI*. Plus and minus signs denote the presence and absence of the bands at a given locus. *HpaII* and *MspI* are isoschizomers that recognize the same tetranucleotide sequence 5'-CCGG, but have differential sensitivity to certain methylation states of the two cytosines: *HpaII* does not cut if either cytosine is fully (both strands) methylated. Whereas *MspI* does not cut if the external cytosine is fully- or hemi- (only one strand) methylated. Thus, for a given DNA sample, unmethylation of cytosines at the 5'-CCGG site result in concomitant presence (+) of the bands for both *HpaII* and *MspI* (case 1). On the other hand, simultaneous full methylation of either all cytosines (case 2) or the external cytosines (marked as CHG, case 3) will be revealed as concomitant absence (-) of a band with both enzymes. Hemimethylation of the external or internal (marked as CG) cytosine (case 4 and 5), as well as full methylation of the internal cytosine (cases 6), is revealed by the presence of a band in only one of the enzyme digests.



Supplementary Fig 2. Examples of MSAP electropherograms on 2x and 4x genotypes of *S. commersonii* generated using *HpaII* and *MspI* isoschizomers, and primers E-TCAA/HM-AGC (*left panel*) and E-TCAA/HM-ACC (*right panel*). *a* and *b*) MSAP traces from *cmm1t* (2x) diploid parent generated using *HpaII*; *c* and *d*) traces of *cmm1t* using *MspI*; *e* and *f*) traces of the synthetic tetraploid *cmm23* generated using *HpaII*; *g* and *h*) traces of *cmm23* using *MspI*. Polymorphic peaks (arrowed in the grey area) show CHG hypermethylation (left panel) and CG hypermethylation (right panel) in *cmm23* respect to its diploid progenitor *cmm1t*.