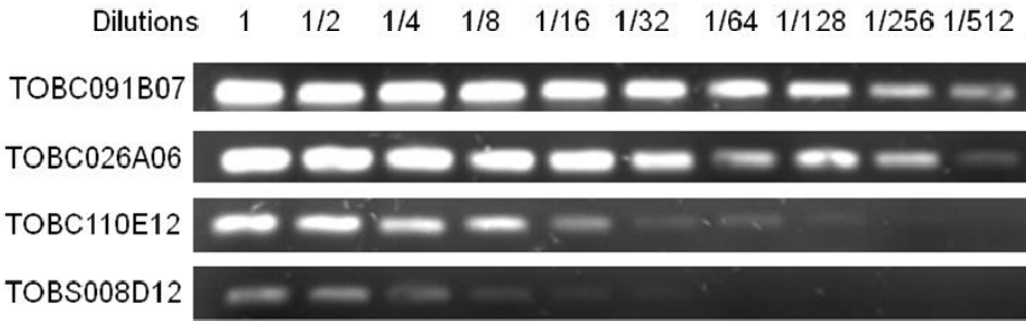

Supplemental Figure S2: Representative ethidium bromide-stained agarose gel (1.5%) of semiquantitative RT-PCR analysis performed from serial dilutions of stigma/style cDNA and primers for the indicated TOBEST sequences. TOBC091B07 cluster, corresponding to TTS1 gene, is composed of 83 clones. TOBC026A06, encoding a non-specific LTP, is composed of 45 clones. The cluster TOBC110E12 (for β-actin), contains 3 independent clones in the TOBEST cDNA library, and TOBS008D12 (for the bZIP transcription factor BZI-3) is a singlet or, in other words, is represented by a unique cDNA clone in the TOBEST.

TOBEST ID	Forward Primer 5'-3'	Reverse Primer 5'-3'	Amplicon Size (bp)
C091B07	TCATGACCACCTTTCACCAGCTCA	AGTAAACAAGGCCACGGACTGCTA	278
C026A06	TTGTGGCGGCGTTAAACGTCTGTT	TCATCAACCTCACTGGACCTTGGA	205
C110E12	TAAGGCTGGATTTGCCGGAGATGA	TCTTCTGGAGCCACACGAAGTTCA	249
S008D12	GATGGCCAACGTTATGCCACAA	TTCTGCCATTTGAGCCCTCAAG	230

Semiquantitative RT–PCR analysis

The stigma/style RNA sample was prepared essentially as described at Materials and Methods for the real time RT-PCR experiments. An aliquot of total RNA (20 μg) was treated with DNase, cleaned with phenol/chloroform, checked for genomic DNA contamination and, after cDNA conversion by reverse transcriptase, the final volume was adjusted to 70 μL with water. For the semiquantitative RT-PCR experiment, an aliquot of 1 μL of the stigma/style cDNA was diluted 100 times. From this dilution, aliquots of 1 μL (equivalent to approximately 2.8ng of total RNA) were considered as point 1 of the semiquantitative RT-PCR experiment. Serial dilutions were performed as

indicated on the figure and, from each dilution, aliquots of 1 µL were taken for the PCR amplifications. The primers used and their amplicon size are indicated on the table above. The PCR conditions were as follows: a first step of 3 minutes at 95ºC, 30 cycles composed of the following three steps, 95ºC for 30 seconds, 55ºC for 30 seconds, and 72ºC for 60 seconds, and a final step at 10°C indefinitely. To exclude the possibility of contamination, samples including all primer pairs but no cDNA were included on the same 96 wells plate (data not shown). These experiments were repeated at least three times with independent replicates and the gel showed at Supplemental Figure S2 is representative of the results found.