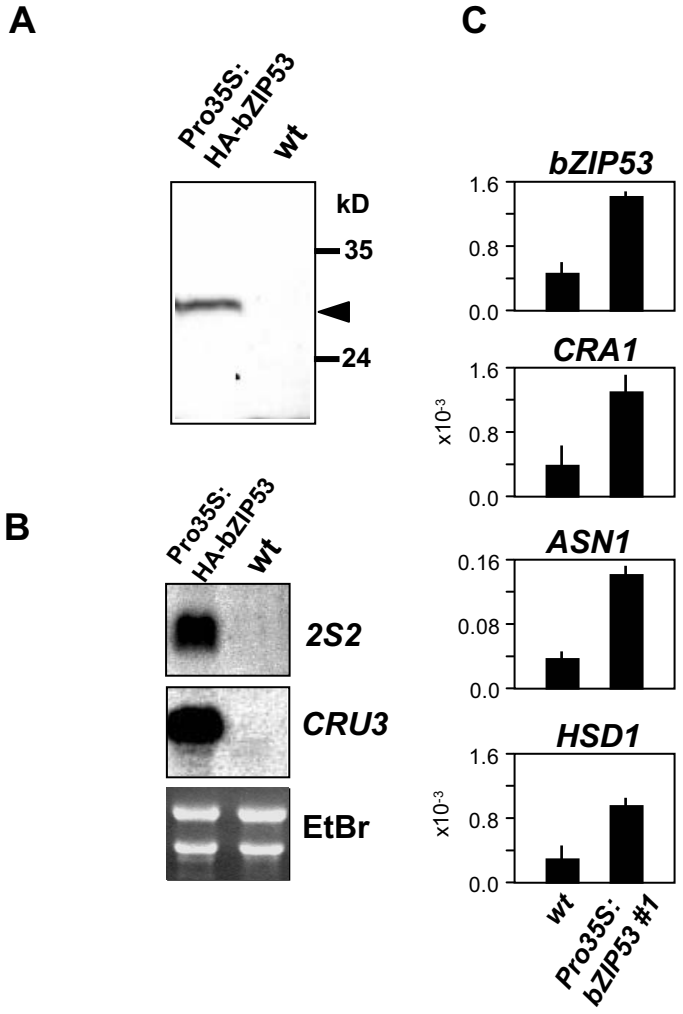
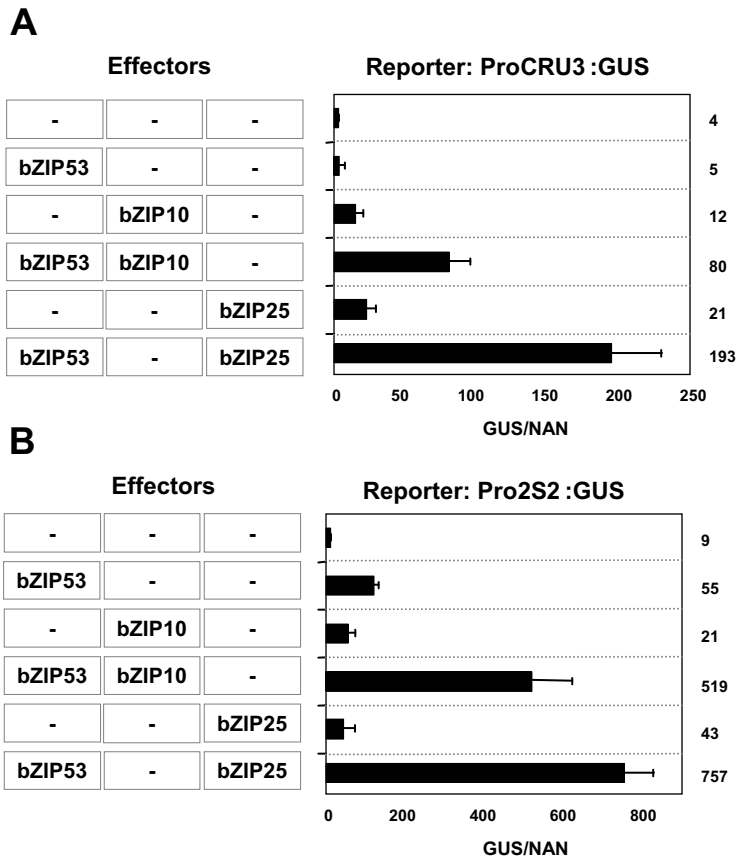
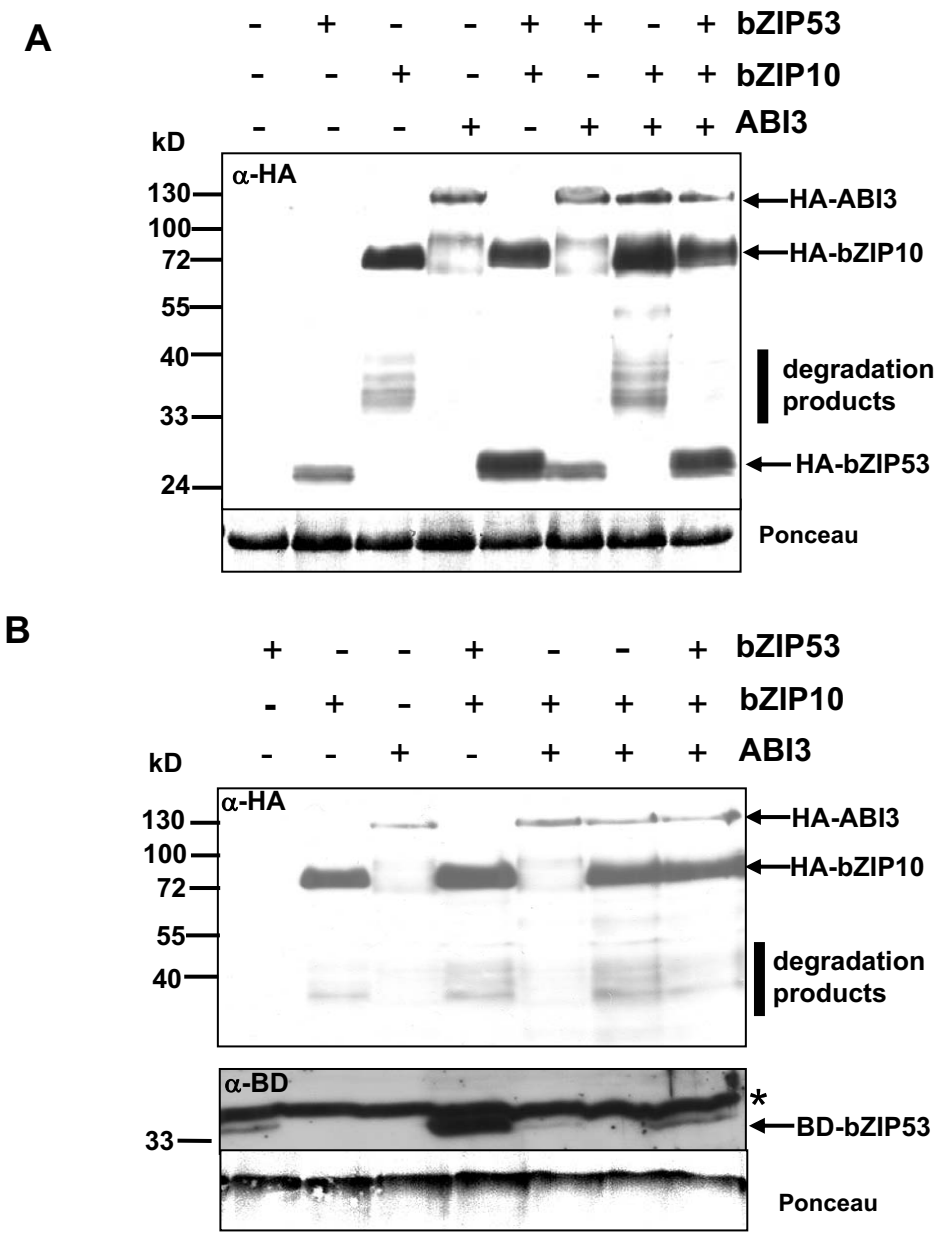
Supplemental Data Alonso et al., (2009) A pivotal role of the basic leucine zipper transcription factor bZIP53 in the regulation of Arabidopsis seed maturation gene expression based on heterodimerisation and protein complex formation.



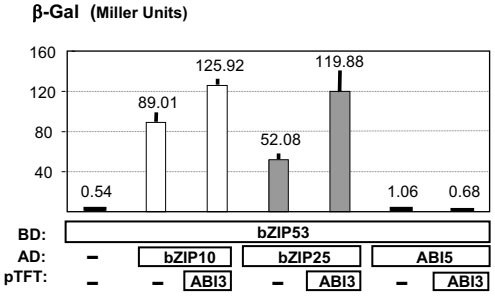
Supplemental Figure 1. Analysis of putative bZIP53 target genes in plants constitutively expressing HA-bZIP53 **(A, B)** or bZIP53 **(C)**. **(A)** Immunoblot analysis of Pro35S:HA-bZIP53 plants using a HA-tag-specific antiserum (α-HA) (arrow). Mass of marker proteins is provided in kilo Dalton (kD). **(B)** Transcription of *2S2* and *CRU3* in leaves of wild-type (wt) and Pro35S:HA-bZIP53 plants has been studied by RNA gel blot analysis. Equal loading was confirmed by Ethidium bromide (EtBr) staining. **(C)** RT-qPCR analysis of two week-old wild-type (wt) and Pro35S:bZIP53 plants (line #1) for expression of *bZIP53*, *CRA1* (Wang et al., 2007), *HSD1* (Li et al., 2007) and *ASN1* (Lam et al., 2003). Expression levels are given as relative to a *UBIQUITIN* (*UBI10*) gene for normalization. Average values and standard errors from four replicates and two different experiments are shown.



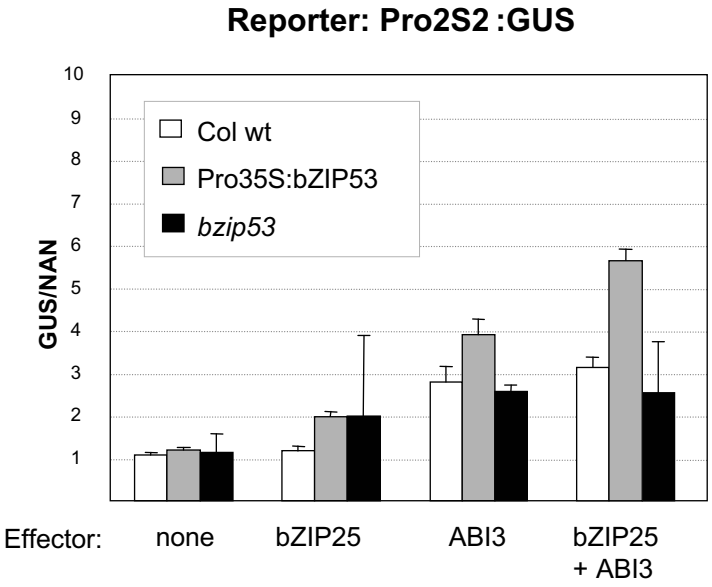
Suplemental Figure 2. Transient expression of bZIP53 in combination with bZIP10 or bZIP25 synergistically enhances transactivation of *CRU3* and *2S2* promoters. Arabidopsis protoplasts were transfected with constructs containing ProCRU3:GUS **(A)** or Pro2S2:GUS **(B)** reporters. Effector constructs containing specific *bZIP* genes under the control of a Pro35S were used in cotransfection experiments. 3 µg of a control plasmid containing a Pro35S:NAN cassette was included in all experiments to normalize GUS expression values for differences in transfection efficiencies as described by Ehlert et al. (2006). X-axis values are expressed as GUS activity relative to NAN activity. Numbers along the Y-axis represent fold-induction values relative to non-transfected control cells. Given are mean values of 4 independent transfections. All experiments were repeated at least 3 times with similar results.



Supplemental Figure 3. Analysis of effector proteins transiently expressed in Arabidopsis protoplasts. Expression of the HA- and Gal4 DNA binding domain-(BD)-tagged bZIP effectors used in the experiments in Supplemental Figure 2 and Figure 6 **(A)** or Figure 4D **(B)**, respectively. Proteins are detected in immunoblot experiments making use of α-HA or α-BD antisera. Protoplasts have been transiently transformed with the constructs indicated. Degradation products and tagged bZIP proteins are marked by a black line or arrows, respectively. Unspecific background staining is indicated by *. Marker proteins are labelled according to their size in kilo Dalton (kD). Equal loading was confirmed by Ponceau staining.



Supplemental Figure 4. Ternary protein interaction studied in a yeast three-hybrid system. Yeast strains (SFY526) expressing different combinations of BD-bZIP53, AD-ABI3 and bZIP10 or bZIP25 were assayed for β-galactosidase activity. The latter were provided in the three-hybrid vector pTFT (Egea-Cortines et al., 1999). Average values (Miller units, M.U.) and standard errors from six replicates and two independent experiments are shown.



Supplemental Figure 5. Transient expression by microparticle bombardment of Arabidopsis leaves from Columbia wild type (Col wt; white bars) and plants with increased (Pro35S:bZIP53; grey bars) or decreased (*bzip53;* black bars) expression of *bZIP53.* Effector constructs containing bZIP25 or ABI3 under the control of a 35S promoter (Pro35S) and a *GUS* reporter gene under the control of an *2S2* promoter were used. X-axis values are expressed as GUS activity relative to NAN activity (Kirby and Kavanagh, 2002). Average values and standard errors from four replicates are shown.

Supplemental Table 1: Oligonucleotide primers used in this study.

***UBI*:**
5´- GCTCTTATCAAAGGACCTTCGG-3´
5´- CGAACTTGAGGAGGTTGCAAAG-3´

bZIP53
5´-TAATGATCCGAGGTACGCCAC-3´
5´-TGCTTCTGTTTCCTCATCCTTG-3´

***CRU3*:**
5´-TAGATGTTCTCCAAGCCACCG-3´
5´-AACGGAAACACCAACACATCG-3´

2S2:
5´-ATTTGCAAGATCCAGCAAGTTG-3´
5'-AATACATTTAGCCTCAAACATC-3'

***LEA76*:**
5´-ACAAAGAGCATTATCCAGGAAGT-3´
5´-ACACAAAGATACTTTCATATCGT-3´

***ProDH*:**
5´-GCATCAAACGGTTCTGGTTTC-3´
5´-TGTTTATCGATCCCGAGGTCA-3´

ASN1:
5´-TTCAACGCCTTATGAGCCTCTT-3´
5´-CACCAGAGAGCAAAACTCCAAA-3´

CRA1:
5´-AGCCCAAATCCAGATCGTAAAC-3´
5´-TCACCACCGAGAAACCTTGTG-3´

***HSD1*:**
5´-TGCCGGAAACAAAGATACGTG-3´
5´-AGTAACCGACAACCCCACTCA-3´