Table S 1

Comparison of priming efficiency of different generations of Ws and *ibs1* lines in response to inoculation with virulent *Pst* (based on data shown in Fig. 3A, B).

| Lines | Priming-factor* in | Priming-factor* in |
|---|----------------------|--------------------|
| | water-treated plants | response to BABA |
| | | treatment |
| WsH / WsB WsB / WsBB WsBH / WsB | 1.4 1.4 1.2 | 1.9 1.6 1.7 |
| ibs1H / ibs1B ibs1B / ibs1BB ibs1BH / ibs1B | 1.2 1.1 1.3 | 1.3 1.1 1.3 |

*The priming-factor was calculated by dividing the bacterial titer in plants of a given generation with the titer in their respective direct descendants. Bacterial titers were determined at 72 h post inoculation.

Table S2: Ct values for the trans-generational lines and treatments at time -48 h

PR1

| line/ | | | | | |
|------------|-------|-------|----------------------|-------|-------|
| treatment | Ct | ± SD | line/ treatment | Ct | ± SD |
| Ws BABA | 19.20 | 0.490 | ibs1 BABA | 18.41 | 1.10 |
| WsB BABA | 18.91 | 0.102 | ibs1B BABA | 17.97 | 0.21 |
| WsBB BABA | 17.88 | 0.015 | ibs1BB BABA | 18.45 | 1.13 |
| WsBH BABA | 19.42 | 0.028 | ibs1BH BABA | 19.67 | 0.14 |
| Ws water | 22.46 | 0.028 | ibs1 water | 24.79 | 0.048 |
| WsB water | 25.43 | 0.007 | ibs1B water | 21.91 | 0.078 |
| WsBB water | 25.58 | 0.070 | ibs1BB water | 22.05 | 0.086 |
| WsBH water | 24.47 | 0.021 | <i>ibs1</i> BH water | 24.52 | 0.021 |
| PR2 | | | | | |
| line/ | | | | | |
| treatment | · | | line/ treatment | | |
| Ws BABA | 19.71 | 0.134 | ibs1 BABA | 18.41 | 0.110 |
| WSB BABA | 19.30 | 0.093 | ibs1B BABA | 17.97 | 0.021 |
| WSBB BABA | 18.71 | 0.042 | ibs1BB BABA | 18.45 | 0.113 |
| WsBH BABA | 19.29 | 0.163 | ibs1BH BABA | 19.67 | 0.014 |
| Ws water | 25.40 | 0.078 | ibs1 water | 24.79 | 0.049 |
| WsB water | 24.63 | 0.049 | ibs1B water | 21.99 | 0.078 |
| WsBB water | 24.68 | 0.255 | ibs1BB water | 22.05 | 0.096 |
| WsBH water | 25.44 | 0.064 | ibs1BH water | 24.52 | 0.021 |
| PR5 | | | | | |
| line/ | | | | | |
| treatment | | | line/ treatment | | |
| Ws BABA | 20.11 | 0.021 | ibs1 BABA | 24.60 | 0.049 |
| WsB BABA | 19.13 | 0.092 | ibs1B BABA | 24.79 | 0.064 |
| WsBB BABA | 19.47 | 0.021 | ibs1BB BABA | 23.96 | 0.015 |
| WsBH BABA | 19.94 | 0.021 | ibs1BH BABA | 23.32 | 0.074 |
| Ws water | 24.11 | 0.049 | ibs1 water | 26.07 | 0.156 |
| WsB water | 25.19 | 0.004 | ibs1B water | 25.23 | 0.219 |
| WsBB water | 24.38 | 0.020 | ibs1BB water | 25.62 | 0.014 |
| WsBH water | 23.58 | 0.060 | <i>ibs1</i> BH water | 25.67 | 0.028 |





Figure S2. *PR1* expression levels in Ws-0 and *ibs1* lines control-treated with water. Fourwk-old plants were treated with water and *PR1* expression was analyzed by qRT-PCR. A, *PR1* expression in Ws-0 lines. B, *PR1* expression in *ibs1* lines. Expression was normalized to the values of WsH and *ibs1*H, respectively, at -48 h. Values represent means +/- SD of three replicates. Similar results were obtained in three independent experiments.

















water

Figure S3. Descendants of BABA-treated Ws-0 plants are more resistant to virulent Pseudomonas syringae. Three-wk-old plants were treated with BABA (25 ppm final concentration in the soil) or water 2 days prior to inoculation with Pst DC3000 (OD₆₀₀ = 0.08). A, Growth of Pst DC3000 in the water-treated Ws-0 lines (WsH, WsB, WsBB and WsBH).B, ibs1 lines (ibs1H, ibs1B, ibs1BB and ibs1BH) at 0, 48 and 72pih. C, Growth of Pst DC3000 in BABA-treated Ws-0 lines compared to that in D. ibs1 lines at 0, 48 and 72 hpi. Bacterial growth was quantified by qRT-PCR as transcript levels of *Psorf* normalized to the transcript level of the Arabidopsis gene AtTUB4. Capital letters indicate statistically significant bacterial growth at 72 h (ANOVA, Student-Newman-Keuls, n = 3, P < 0.05). Small letters indicate statistically significant bacterial growth at 48 h (ANOVA, Student-Newman-Keuls, n = 3, P < 0.05). E, Disease response of the BABA- or water-treated of ibs1 lines at 5 days postinoculation. Small and capital letters above error bars indicate statistically significant differences in the percentage of leaves with symptoms in BABA- and water-treated lines, respectively (ANOVA, Student-Newman-Keuls, n = 30, P < 0.001). F, Visible disease phenotype in the Ws-0 lines. G, Visible disease phenotype in the *ibs1* lines.



Figure S4. qRT-PCR analysis of *PR2* and *PR5* transcript levels in Ws -0 and *ibs1* lines upon inoculation with virulent *Pst*. Expression levels of PR2 (A) and PR5 (B) in BABA - treated (circles) and water -treated (squares) Ws -0 and *ibs1* lines. Expression was normalized to the corresponding sample treated with water at 0 h. The values represent means +/- SD of three replicates. Similar results were obtained in three indepe ndent experiments.

А

В



Figure S5. qRT-PCR analysis of *PR2* and *PR5* transcript levels in Col -0 lines upon inoculation with virulent *Pst*. Expression levels of *PR2* (A) and *PR5* (B). Expression was normalized to the corresponding sample at 0 h. The values represent to means +/- SD of three replicates. Similar results were obtained in three independent experiments.



Figure S6. Priming of the progeny of BABA -treated lines is not due to direct transfer of BABA to the next generation. BABA concentrations were determined in u ntreated plants and in plants soil-drenched with BABA, respectively. A,BABA 40 ppm and B,BABA 25 ppm. S_1 = first generation selfed progeny. The horizontal lines at 50 µg BABA/g dw show the threshold concentration for resistance induction.



Figure S7. Graphical output generated by CyMate of the Ws transgenerational lines.

- A) Methylation analysis of a 503 bp fragment of the *PR1* promotor of Ws and descendants. The location of the cytosines within the sequenced fragment is shown in the ruler at the top of the Figure. Beneath the ruler, the filled symbols represent methylated cytosine residues, whereas the open symbols represent cytosine residues lacking methylation. The red circles stand for ^mCGN, the blue squares represent ^mCHG and the green triangles represent ^mCHH.. The uppermost sequence (Ws-0) corresponds to consensus sequence and is followed by individual clones obtained by polymerase chain reaction amplification of bisulfite-treated DNA. The order of the individual sequences is shown on the left.
- B) Zoom-in view of the *PR1* promotor fragment showing the different motives (WRKY, TGA, ERF, PR-box and NFκB) identified on the promotor.

| ruler | 1 | | | | | | | 1 | 1 | | | | | | 1 | 1 | | | | | | 15 | 1 | | | | | | 20 | 1 | | | | | | 25 | 1 | | | | | - | 301 | | | | | | 35 | 1 | | | | | 40 | 1 | | | | | 4 | 51 | _ | _ | | | _ | 39 | P ³ |
|--------------------------|---|----|---|----------|----|-----|-----|-----|--------|--------|-----|-----|-----|-----|-----|--------|-----|-------|-----|-------|-------|-------|------------------|------------------|-------|------|--------|------|-----------|----|----|----|----|----|----|------------------|-------|-----|-----|-----|-----|-----|-----|-----|-----|----|---|----|----|------------------|-----|----|------|----|----|-----|-----|-----|-----|---|----------|----|----|----------|---|-----|-----|-----|-----------------------|
| Ws-0 ibs1H1 ibs1H2 | | // | | / | / | | | | | | | | " | | | 1/1/1- | | - /// | | T//// | 1/1/1 | 1//// | | 1//// | 1//// | //// | - //// | //// | · / //*** | // | // | // | / | | | - WILLANA | -//// | | | /// | | | | / | 1 | | | - | / | | | // | 7/10 | | 1 | • | | | | | | | // | // | | 7/× | × | | |
| ibs1H3 | 2 | 2 | 1 | 1 | | | | • | | | | • | | | | 2 | | | . 2 | 1 | 2 | 1 | | 1 | t | E. | t | P | Ł | 2 | 2 | 2 | | | | 2 | 2 | _ | - 5 | 2 | : | - | 2 | | | | 2 | 2 | | Ρ. | | 2 | P | P | 2 | _ | | | | 2 | 2 | 2 | Ł | 2 | 2 | | | . ! | 2 |
| inclus | 5 | 5 | 5 | 1 | | | | | | | | | | | | | 1 | | | 5 | . 5 | 1 | K | 5 | 5 | 2 | 2 | 5 | 5 | 5 | 2 | 5 | | | 5 | 5 | 5 | | | | | ÷ | | | Ľ | | 5 | 5 | 5 | | - | 5 | 1 | 5 | 5 | ~ | | | | 5 | 5 | 5 | 5 | 5 | 5 | | - 2 | | 5 |
| ibs1H6 | | - | | 2 | 1 | | | | | | | - 1 | | | 5 | | 1 | | 1 | ~ | | | | - | | | D | 5 | | 5 | ō | b | Ĕ. | Ξ. | 5 | 5 | | | | | | ÷ | 1 | | | | | | | | | 1. | D | | Ď | ŏ | | | | | | | | 5 | | | - 7 | • • | |
| ibs1H7 | Þ | • | | Ē | i | - 1 | - | • | - | Þ i | - | 0 | > (| 0 | >) | | Þ | | • | Þ | | | | Þ | | | | | D | | | | | ō | Þ | Þ | | | | 0 0 | 0 | | • | | D | Þ | | | | DI | | • | Þ | | | õ i | | | | | | | | | | | | • 1 | |
| ibs1H8 | | | | | | - 1 | - | • | • | • | • | • 1 | • | | > > | • | | | Þ | Þ | Þ | | • | ٠ | ۲ | ۲ | • | ۲ | > | ۲ | | Þ | | | • | • | • | | | • | • • | | | • • | Þ | | ۲ | ۲ | • | - | | • | | | • | 0 | • • | ·P | | ۲ | ۲ | ۲ | | ۲ | ۲ | | | | • |
| ibs1H9 | • | • | • | | | - 1 | • | 0 | • | | > | 0 | • | • • | • • | • | • | | Þ | | • | • | • | ۲ | ۲ | ۲ | ۲ | ۲ | ۲ | ۲ | | ۲ | | | • | • | • | | | • • | • • | | | • • | Þ | | ۲ | ۰. | • | • 1 | • 0 | • | • | ٠ | • | 0 | • • | • • | Þ | Þ | ۲ | ۲ | • | ۲ | ۲ | | | • • | * |
| ibs1sH10 | | • | | | | | | • | | | | • | > | P | > • | | | | | | 1 | P | P | P | • | • | • | | • | | | • | | | P | | | -1 | | • 1 | | | • | • • | P | | | • | | | | | • | • | | 0 | • • | • • | | | • | P | • | • | • | | | • • | • |
| 1051H11 | 2 | 2 | | | | > 1 | 2 | 0 | 2 | 2 | 2 | 0 | > | | | 2 | | ۰. | | 2 | P | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 | P | | t. | 2 | 2 | 2 | 2 | 2 | | | 2 | 0 | - | | | | P | P | P | | P | 2 | 1 | 1 | 2 | 2 | 0 | 2 ! | | | 2 | 2 | 2 | 2 | 2 | 2 | | | | 2 |
| ibs1H12 | 1 | 5 | | 2 | | | | | | | | 5 | | | | - | | - | | | 1 | 1 | 5 | 5 | 2 | 5 | 5 | 5 | 2 | 5 | - | 5 | - | - | 5 | 5 | 5 | - | | | | ÷ | | | | 1 | 5 | 2 | 2 | | | | | 5 | 5 | 2 | | | | 5 | 5 | 5 | 5 | 5 | 5 | | | | 5 |
| ibs1H14 | 5 | 5 | | ÷ | ł. | | | | | | | ŏ | | | | 9 | 15 | | | | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | | 5 | - | Π. | 5 | 5 | 5 | - | | ŏ | | ÷ | 6 | | | | - | | | 5 | | 5 | | 5 | 5 | ŏ | | | 6 | | 5 | 5 | 5 | 5 | 5 | | - 7 | | 5 |
| ibs1H15 | | | | | | - | | | - | | - | ŏ | | | | | 1 | | | | | | | | | | | | Ď | | - | | Ξ. | ŏ | | | | | | ŏ | | | Ď | | | | | | | 51 | | | | | | ŏ | | | · D | | | | | | | | - 7 | ē i | |
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| ibs184 | P | R | | | | 2 | 2 | | 2 | P | | : : | | ١. | | 4 | | | | - | P | P | P | 2 | 2 | 2 | 2 | 2 | 2 | 2 | - | t | H | - | 2 | 2 | 2 | | | 2 2 | | ÷ | | | | 2 | 2 | 2 | 2 | | 4 | | | 2 | 2 | 2 | | | | 2 | 2 | P | 2 | 2 | 2 | - | . : | | 2 |
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| 05189 | • | | • | . 0 | 2 | | | • | • | | | • | - | • • | >) | | | | P | P | P | • | • | • | ۲ | ۲ | • | | • | ۲ | | • | | | • | • | • | | | 0 | 0 | | • | ·P | • • | • | | ۲ | • | | | Þ | • | | • | 0 | • • | • • | • | • | • | • | • | ۲ | • | _ | • • | • • | • |
| Det 014 | P | P | | | | | 2 | 0 | 2 | | | • | 2 | | | 2 | 1 | | P | | 1 | 2 | 2 | 1 | 2 | 2 | 2 | 2 | 2 | 2 | - | P | 0 | 0 | 2 | 2 | 2 | | - 1 | • | • | | | | | 1 | 2 | 2 | 2 | | 2 | P | 1 | P | 2 | 0 | | | | 2 | 2 | 2 | 2 | 2 | 2 | - | | . ! | 2 |
| ibs1812 | 1 | 5 | | ÷ | 4 | | | | 5 | | | | | | | | | | | | | 1 | 5 | 5 | 5 | 5 | 2 | 2 | 5 | 5 | - | 5 | • | 2 | 5 | ٢. | 5 | | | | | 4 | | | | | 5 | ٢. | 5 | | | | | 5 | 5 | 8 | | | | 5 | 5 | 5 | 5 | 5 | 5 | - | | | 2 |
| ibs1813 | | 5 | | | | | | | | | | | | | | | 1 | i i | Ь | 5 | D. | | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | E | 5 | - | | 5 | 5 | 5 | | 5 | 0 | ō | | 1 | D | | | 5 | 5 | 5 | | | Ь | | | 5 | ŏ | | | | 5 | 5 | 5 | 5 | 5 | 5 | | | - 1 | |
| ibs1814 | | | | | | - | | • | | | | • | - | | | | 5 | | 1. | | | | | | | | | | | | = | | F | Ξ. | | | | - 1 | | | ě | | | | | | | | | | | 1 | | | | DI | | | | | | | | | | Ξ. | | ē i | |
| ibs1815 | • | | | | | • 1 | • | • | • | • | • | 0 | • | • • | | | • | | • | | | | | | ۲ | | • | ۲ | ۲ | | | ۲ | | | • | • | • | | | • | • (| | | | | | ۲ | • | • | - | • | • | Þ | | • | 0 | | | | Þ | ۲ | • | Þ | ۲ | | | • • | • 1 | • |
| bs1816 | • | • | • | · C |] | - 1 | - | • | • | | | 0 | • | • • | • • | | | | | | | Þ | | P | | | • | ⊳ | • | • | | • | | | • | • | • | | | 0 | 0 | . 0 | • | ·P | • • | • | ۲ | • | > | | | Þ | | • | • | 0 | • • | • • | • | • | • | | • | ۲ | ۲ | • | • • | • • | • |
| 051881 | | | | 5 | | | - | 0 | \geq | \geq | | • | > | | | | | - 5 | P | P | P | 1 | 2 | P | | 2 | • | • | P | 1 | | 2 | | | P | P | P | | | 0 0 | 0 | | 2.2 | ·P | | 1 | P | | 2 | | 2 | P | | | • | 0 | | | | P | 2 | P | P | 2 | | - | • | • ! | 2 |
| ibs1883 | 1 | 2 | | - | | | | | | | | 0 | | • • | 2 | | | - | P | P | 2 | P | P | 2 | 2 | 2 | 2 | 2 | 2 | 2 | - | 2 | 2 | Ч. | 2 | 2 | 2 | - | | | • | ٠ | | | | P | 2 | 2 | 2 | 2 | | | | 2 | 2 | 2 | | P | | 2 | 2 | 2 | 2 | 2 | 2 | - | | | 2 |
| ibs1884 | 5 | 5 | 5 | ÷ | | | | | | | | Ξ. | | . : | | | 5 | | | | | 5 | 5 | 5 | 5 | 5 | 5 | 5 | ς. | 5 | - | 5 | - | - | 5 | 5 | 5 | - | | Ξ, | ~ | • | | | | | 5 | 5 | 5 | 5 | | | ~ | 5 | 5 | 2 | | | | 5 | 5 | 5 | 5 | 5 | 5 | - | = 7 | | 5 |
| ibs1885 | 5 | - | 5 | ŀ | e. | | | | | | | | | 5 | | | 1 | | 5 | 5 | | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | = | 5 | - | - | 5 | 5 | 5 | - | | ŏ, | | ÷ | š | | | | 5 | 5 | 5 | 5 | | 16 | 5 | 5 | 5 | ŏ | | | | 5 | 5 | 5 | 5 | 5 | 5 | - | | | 5 |
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| ibs18812 | • | ► | • | | | - 1 | - | • | > | | • | • • | • | Þ | > > | | | | | • | • | | | | ► | ۲ | ۲ | ۲ | ۲ | ۲ | | ۲ | | | ۰. | \triangleright | | | | • | • | | • • | • • | • • | ٠ | | | • | • • | > C | • | | • | • | 0 | Þ D | > ► | • | • | ۲ | ۲ | • | • | ۲ | | • • | • • | ٠. |
| ibs18H1 | Þ | | | | 0 | • | • | • | | | - | • • | • • | | • • | | | | | • | P | • | Þ | • | • | • | | | • | • | | Þ | | | ۰. | | • | | | • | • | | P | > > | • | • | • | • | • | • • | | Þ | • | • | • | 0 | • • | • P | • • | | • | ۲ | • | ۲ | ۲ | | • • | • • | ٠. |
| ibs18HZ | ۲ | • | • | | • | · P | > (| 0 | • • | - 1 | • (| O D | > (| D D | • • | • • | • | | ► | • | • | | ۲ | ۲ | • | • | • | ► | • | ۲ | | ۲ | | | | | • | | • | • | • | | D | > Þ | · Þ | • | • | • | | \triangleright | | | Þ | | Þ | 0 | • • | • • | ·Þ | • | • | | • | ۲ | • | | • • | • • | • |
| IDS18H3 | 1 | 1 | ۲ | | P | r D | > | | | • | > (| D D | > (|) Þ | • • | | | | Þ | • | 1 | 1 | | • | t | 2 | 2 | ۲ | 2 | | | 2 | | | | | | | | 0 0 | 0 | | | | 1 | P | 1 | P | | PI | | Þ | P | P | P | 0 | 2 9 | P | | 1 | P | P | P | 1 | | | • • | • ! | 2 |
| IDS18H4 | 2 | 2 | | | | | 5 | 2 | | | - | | 2 | | | 2 | | | | | 1 | P | 2 | | 2 | 2 | 2 | | 2 | | | 2 | | | | | | | | | | | P | P | P | 1 | 2 | 2 | • | 2 | | | P | P | P | 0 | | | P | 1 | 2 | P | 2 | 2 | | | : 1 | | 5 |
| ibs18H6 | 2 | 5 | 5 | | | | | 2 | ۰. | | | | ٢, | | | | | - | | | 1 | 2 | 5 | 2 | 5 | 2 | 2 | 5 | 5 | 2 | = | 5 | - | | 5 | - | 5 | | | | | | 2 | R | K | 1 | 2 | 5 | | 2 | | 1 | K | K | 2 | 2 | | | K | 1 | 5 | K | 5 | 0 | 5 | - | | | 5 |
| ibs1BH7 | 5 | 5 | 5 | | 1 | | | 1 | | | | | | | 1 | | 1 | - | | | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 2 | 5 | Ξ. | 2 | 5 | - | 5 | | | | | | K | | | 1 | 5 | 5 | 5 | | | 1 | 1 | 5 | 5 | 8 | | | | 5 | 5 | 5 | 5 | 5 | 5 | - | - 1 | | 5 |
| ibs18H8 | 5 | 5 | 5 | F | 1 | | | 5 | | | | | 1 | 5 | | | 1 | F | 5 | 5 | 5 | D | 5 | | 5 | 5 | 5 | 5 | 5 | 5 | 1 | 5 | 2 | 6 | 5 | | 5 | | | | | | Б | 6 | | 5 | 5 | 5 | | 6 | | 1 | 5 | b | Ď | ŏ | | | 6 | 5 | - | D | 5 | 5 | 5 | | | | |
| | | | | - | | | _ | | | | | | | | - | - | | | | | | | | | | | | | | | Ξ. | | | | | | | | | | | | - | - | - | | | | | | | | | | | | | _ | - | | <u> </u> | - | | <u></u> | - | | | | |
| position | | 2 | 3 | 0 | 3 | 2 | 0 | 2 | 0 | 200 | 0 | 78 | 7 | 2 | 8 | 5 | 8 | 3 | 8 | 0 | 8 | B | 3 | B | 3 | 8 | P | S | 0, | 8 | 3 | 3 | 3 | 3 | 30 | 91 | 3 | p | 5 | 24 | Bq | BR | 38 | 18 | 3 | 10 | 3 | 3 | 20 | 8 | 1 1 | 18 | B | S. | B | 3 | 53 | B | B | 3 | 2 | 3 | 3 | B | 2 | 8 | 28 | 30 | 3 |

Figure S8. Graphical output generated by CyMate of the *ibs1* trans-generational lines. Methylation analysis of 503 bp fragment of *PR1* promotor of *ibs1* and descendants.Filled symbols represent cytosine methylation, wheras open symbols represent lack of methylation. Circles represent ^mCG, squares represent ^mCHG and triangles represent ^mCHH. The order of the individual sequences is shown on the left.



Fig. S9: Bilsulfite sequencing data showing percentage of CGN, CHG and CHH methylation, as computed by the CyMate program, in a 503-bp region of the *PR1* promotor of Ws-0 and descendants and *ibs1* and descendants, respectively.