

**Supplemental Figure 1: Confirmation of higher-order mutants using root growth assays or expression analysis.** A-B) Combination mutants and wild type were grown on no hormone, 20  $\mu$ M IBA (A) or 80  $\mu$ M IAA-Ala (B) and root length was measured after 10 days.  $n \geq 15$ ; error bars represent standard error. Asterisks indicate statistically significant changes between the wild type and mutant on media supplemented with hormone by Student's t test ( $p < 0.05$ ). C) *MES17* expression was measured using quantitative PCR to confirm the *mes17 ibr3* mutant. Asterisk indicates a statistically significant difference by Student's t test ( $p < 0.05$ ).

**Supplemental Figure 2: Germination changes resulting from alterations in auxin homeostasis are strongest following disruption to the IBA response and IAA conjugate hydrolase pathways.** A) IAA rescues the germination defect seen in the *ill2 ilr1 iar3 ibr1* mutant. B) Wild-type germination on day 3 is similar under all the conditions tested. C) *ill2 ilr1 iar3 ibr1* germination delays after 3 days are rescued by IAA but not IBA, IAA-Ala, or IAA-Leu. D-E) Combinatorial mutants with defects in *MES17* show weak defects in germination on day 3. Letters indicate significant differences using one-way ANOVA ( $P < 0.05$ ).  $n=3$ ; error bars represent the standard error.

**Supplemental Figure 3: Combination mutant root defects are rescued by application of exogenous auxin.** A) Combination mutants and wild type were grown on media containing 20 nM IAA and root length was measured.  $n \geq 15$ ; error bars represent standard error. No significant differences were detected ( $P < 0.05$ ). B-C) For lateral root measurements, mutants and wild type were grown on media containing no hormone for 4 days and transferred to media without hormone (shown in B and C using a different y axis) or containing the synthetic auxin NAA (C). The number of lateral roots and primary root length was measured after 14 days. Root density is the number of lateral roots per millimeter of primary root length.  $n \geq 15$ ; error bars represent standard error. Asterisks indicate statistically significant results by Student's t test ( $p < 0.05$ ).

**Supplemental Figure 4: Combination mutants show normal auxin levels in growing tissues.** Auxin levels were measured from seedling meristem and hypocotyl (A, B) and cotyledon tissues (C). Auxin levels were measured as in Figure 5. No significant differences are seen between samples using one-way ANOVA ( $P < 0.05$ ).

**Supplemental Figure 5: Combination mutants have normal gravitropic responses.** Wild type and combination mutants were grown on media containing no hormone in the light for 4 days. The plates were then turned 90° and the roots were grown for an additional 5 days in the dark. One representative individual from each line is shown.