

Forward genetic screen for auxin-deficient mutants by cytokinin

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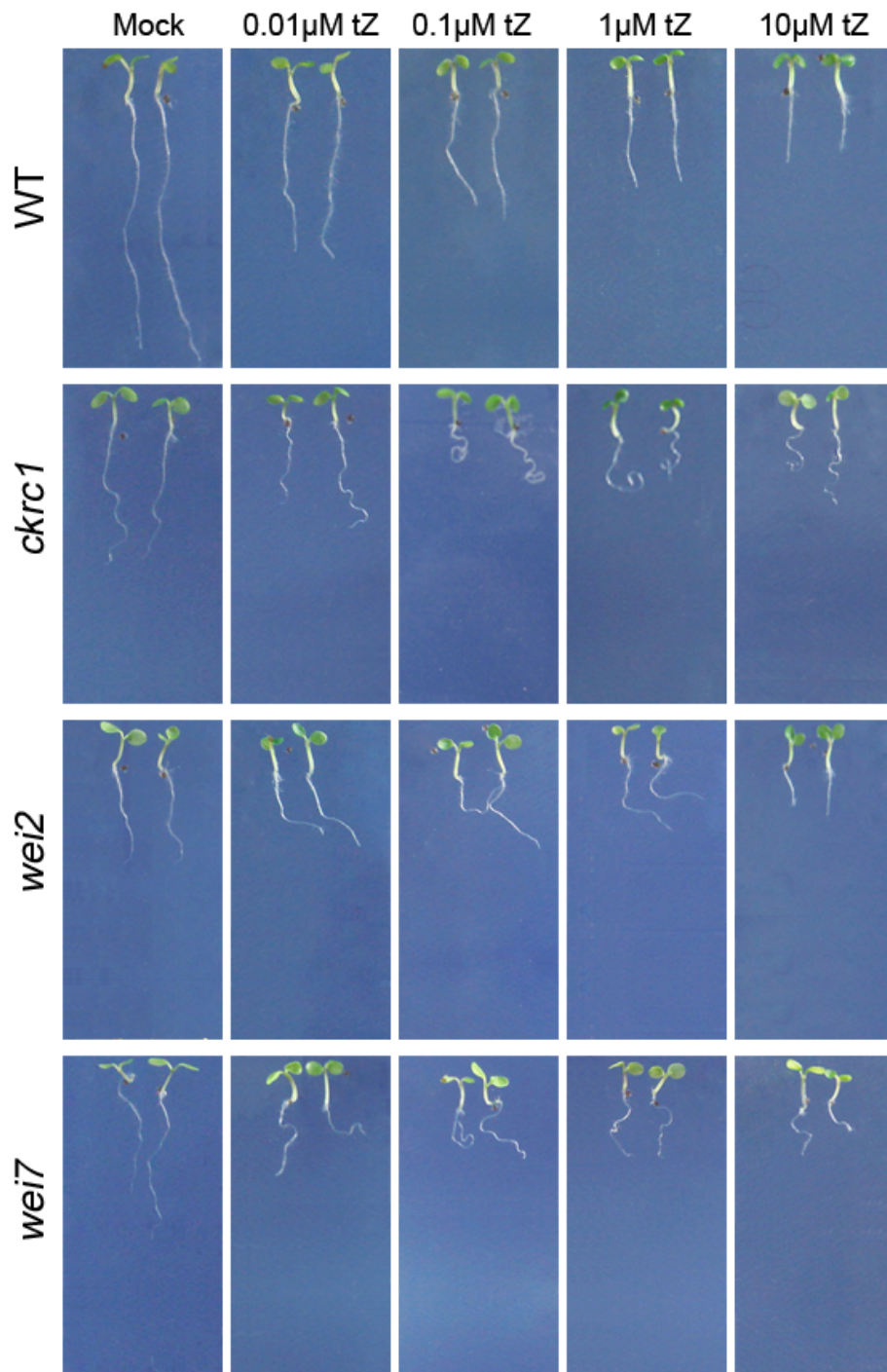
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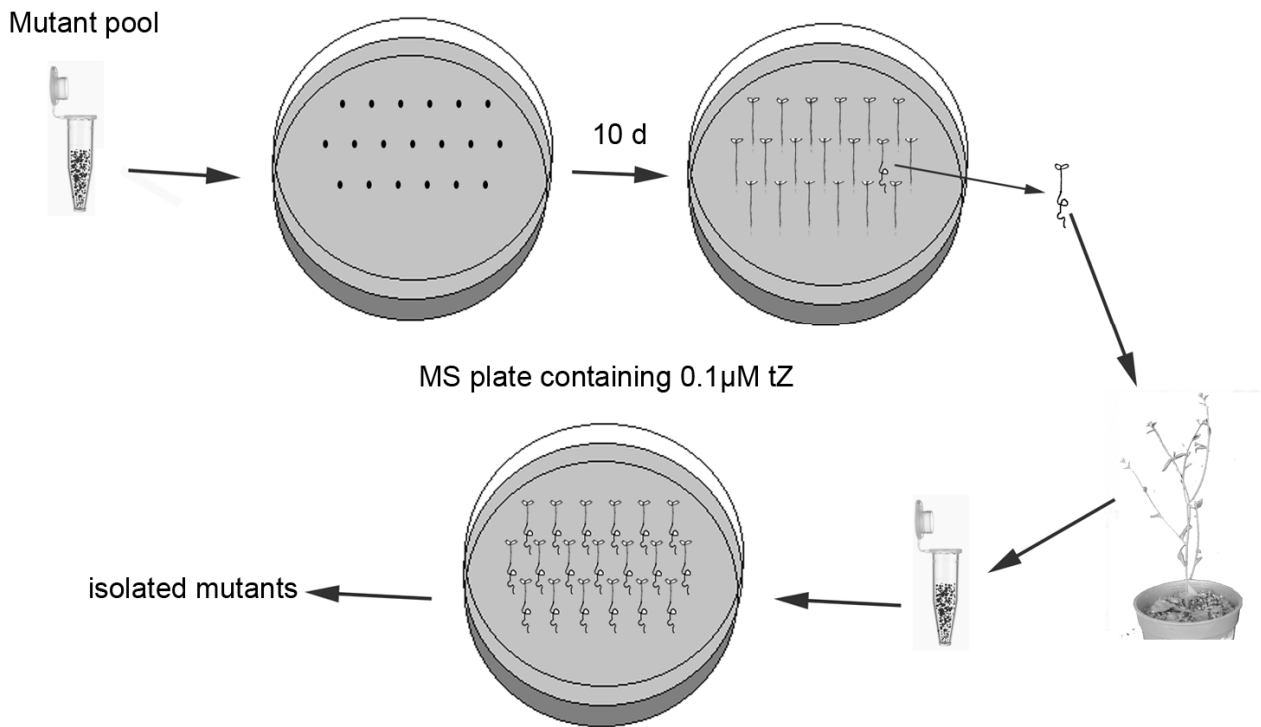
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The author(s) declare no competing financial interests.



Supplementary Fig. S1 Root phenotypes of WT, *ckrc1-1*, *wei2* and *wei7* seedlings on MS plates with indicated cytokinin concentrations 7 DAG.



Supplementary Fig. S2 Schematic diagram of the two-round forward genetic screen for mutants using CK-induced root curling/waving as a phenotypic marker.

Supplementary Appendix S1 PCR primer sequences

QRT-ARR5-F: CCAGTCATCCCAGGCATAGAGT

QRT-ARR5-R: GATCGGAAGTTCATCGAGCGG

QRT-ARR15-F: GGAGTGTCGTCATCAAGGGAG

QRT-ARR15-R: GAGGTGGTGAAGCTGAAGAAGG

QRT-ACT8-F: TGTGACAATGGTACTGGAATGG

QRT-ACT8-R: TTGGATTGTGCTTCATCACC

QRT-IAA1F(2): CGGTTAGATCTCACTGGAGGCCAT

QRT-IAA1R(2): ATCTGCTCCTCCTCCTGCAAAAAC

QRT-IAA2F: AGGAAGAGTCTAGAGCAGGAGC

QRT-IAA2R: ACTGGATGTTGGTTGGTGATG

Supplementary Table S1 Summary information about the 53 mutants with curling (rc) or waving (rw) roots following tZ exposure obtained in the first large-scale screen.

| Mutant | T-DNA vector | Dominant/recessive(r) | Flanking sequence | Allele | Group |
|---------------------|---------------------|------------------------------|---|----------------------|--------------|
| <i>rc1</i> | pROK2 | r | AT2g39990 | <i>pin2</i> | I |
| <i>rc2</i> | pROK2 | r | | <i>pin2</i> | I |
| <i>rc3</i> | pROK2 | r | AT2g38120 (AUX1) | <i>aux1</i> | I |
| <i>rc4</i> | pROK2 | r | AT1g70560 (CKRC1) | <i>ckrc1</i> | II |
| <i>rc5</i> | pROK2 | r | AT1g70560 (CKRC1) | <i>ckrc1</i> | II |
| <i>rc6</i> | pROK2 | r | AT1g01650; AT4g21903; AT5g57090 (PIN2) | <i>pin2</i> | I |
| <i>rc7</i> | pROK2 | r | AT2g43140-AT2g43150; AT2g38120 (AUX1) | <i>aux1</i> | I |
| <i>rc8</i> | pROK2 | r | AT1g53930-AT1g53935; AT1g70560 (CKRC1) | <i>ckrc1</i> | II |
| <i>rc9</i> | pROK2 | r | AT2g38120 (AUX1) | <i>aux1</i> | I |
| <i>rc10</i> | pROK2 | r | AT2g11740; AT1g53930-AT1g53935; AT2g47130-AT2g47140 | <i>pin2</i> | I |
| <i>rc11</i> | pROK2 | r | AT1g61940-AT1g61950 | <i>aux1</i> | I |
| <i>rc12</i> | pSKI015 | r | | <i>aux1</i> | I |
| <i>rc13</i> | pSKI015 | r | AT4g27745 | <i>wei7</i> | II |
| <i>rc14</i> | pSKI015 | r | AT4g27745 | <i>ckrc1</i> | II |
| <i>rc15</i> | pSKI015 | r | AT4g27745; AT1g70560 (CKRC1) | <i>ckrc1</i> | II |
| <i>rc16</i> | pSKI015 | r | AT1g70560 (CKRC1) | <i>ckrc1</i> | II |
| <i>rc17</i> | pSKI015 | r | AT5g04340-AT5g04347 | <i>aux1</i> | I |
| <i>rc18</i> | PROK2 | r | AT1g61940-AT1g61950 | <i>aux1</i> | I |
| <i>rc19</i> | pSKI015 | r | AT1g70560 (CKRC1) | <i>ckrc1</i> | II |
| <i>rc20</i> | pSKI015 | r | AT1g70560 (CKRC1) | <i>ckrc1</i> | II |
| <i>rc21</i> | pSKI015 | r | AT2g38120 (AUX1) | <i>aux1</i> | I |
| <i>rc22</i> | pSKI015 | r | AT2g17390; AT1g70560 (CKRC1) | <i>ckrc1</i> | II |
| <i>rc23</i> | pSKI015 | r | AT3g55210-AT3g55220; AT2g38120 (AUX1) | <i>aux1</i> | I |
| <i>rc24</i> | pSKI015 | r | AT2g46950 (CYP709B2) | <i>wei2</i> | II |
| <i>rc25</i> | pSKI015 | r | AT3g55210-AT3g55220 | <i>aux1</i> | I |
| <i>rc26*</i> | pSKI015 | r | | <i>ckrc2*</i> | II |
| <i>rc27</i> | pSKI015 | r | AT4g01260-AT4g01265 | <i>aux1</i> | I |
| <i>rc28</i> | pSKI015 | r | AT1g71280-AT1g71290; AT5g48000-AT5g48010 | <i>aux1</i> | I |
| <i>rc29</i> | pSKI015 | r | AT2g38120 (AUX1) | <i>aux1</i> | I |
| <i>rc30</i> | pSKI015 | r | AT5g57090 (PIN2) | <i>pin2</i> | I |
| <i>rc31</i> | pSKI015 | r | AT2g14450-AT2g14455; AT2g17930 | <i>aux1</i> | I |
| <i>rc32</i> | pSKI015 | r | AT3g55210-AT3g55220; AT2g38120 (AUX1) | <i>aux1</i> | I |
| <i>rc33</i> | pSKI015 | r | AT3g51860-AT3g51870 | <i>aux1</i> | I |
| <i>rc34</i> | pSKI015 | r | | <i>pin2</i> | I |
| <i>rw2*</i> | pSKI015 | r | AT5g25425-AT5g25430 | <i>ckrw2*</i> | III |

| | | | | | | | |
|--------------------|----------------|----------|---|--|------------|----------------------|------------|
| <i>rc36</i> | pSKI015 | r | | | | <i>wei7</i> | II |
| <i>rc37</i> | pSKI015 | r | | | | <i>aux1</i> | I |
| <i>rc38</i> | pSKI015 | r | | | | <i>pin2</i> | I |
| <i>rw1*</i> | pSKI015 | r | AT5g49665 (<i>WAV3</i>); AT4g18430; AT4g08090 | | | <i>ckrw1*</i> | III |
| <i>rc40</i> | pSKI015 | r | | | | <i>aux1</i> | I |
| <i>rc41</i> | pSKI015 | r | | | | <i>ckrc1</i> | II |
| <i>rc42</i> | pSKI015 | r | | | | <i>aux1</i> | I |
| <i>rc43</i> | pSKI015 | r | | | | <i>wei7</i> | II |
| <i>rc44</i> | pSKI015 | r | | | | <i>pin2</i> | I |
| <i>rc45</i> | pSKI015 | r | | | | <i>pin2</i> | I |
| <i>rc46</i> | pSKI015 | r | AT1g15680-AT1g15690; AT2g14450-AT2g14455 | | AT1g15780; | <i>pin2</i> | I |
| <i>rc47</i> | pSKI015 | r | AT4g27745; AT1g71280-AT1g71290; AT1g15780 | | | <i>wei7</i> | II |
| <i>rc48</i> | pSKI015 | r | AT2g14450-AT2g14455; AT5g06610 | | | <i>wei7</i> | II |
| <i>rc49</i> | pSKI015 | r | AT2g14450-AT2g14455; AT1g68820-AT1g68825; AT1G70560 (CKRC1) | | | <i>ckrc1</i> | II |
| <i>rc50</i> | pSKI015 | r | | | | <i>wei2</i> | II |
| <i>rc51</i> | pSKI015 | r | AT2g14450-AT2g14455 | | | <i>aux1</i> | I |
| <i>rc52</i> | pSKI015 | r | AT5g14910-AT5g14920; AT2g14450-AT2g14455; AT1g70560 (CKRC1) | | | <i>ckrc1</i> | II |
| <i>rc53</i> | pSKI015 | r | | | | <i>ckrc1</i> | II |

Supplementary Table S2 Summary information about the 96 mutants with curling or waving roots following tZ exposure obtained in the subsequent large-scale screen.

| Pools | Vector | Ecotype | No. of lines | No. seedlings screened | No. isolated mutants | No. confirmed known alleles | New rc/rw mutants | Group |
|----------|-----------|---------|--------------|------------------------|----------------------|--|---|---------------|
| CS 31400 | pSKI015 | C24 | ~9,700 | ~24,700 | 2 | | | |
| CS 31402 | pSKI015 | C24 | ~4,000 | ~10,320 | 1 | 1 <i>aux1</i> | | I |
| CS 21991 | pSKI015 | Col-7 | ~8,200 | ~108,810 | 6 | 3 <i>aux1</i> , 2 <i>pin2</i> | | I I |
| CS 21995 | pSKI015 | Col-7 | ~8,600 | ~48,860 | 1 | | | |
| CS 23153 | pSKI015 | Col-7 | ~6,200 | ~63,900 | 16 | 1 <i>ckrc1</i> , 5 <i>pin2</i> , 1 <i>wei2</i> , | <i>ckrc3(ddw41)</i> | II I II |
| CS 31087 | pD991 | Col-6 | ~11,300 | ~113,060 | 16 | 2 <i>ckrc1</i> , 9 <i>aux1</i> , 1 <i>pin2</i> | <i>ckrc4(zb11)</i> <i>crcl(le10)</i> | II I I |
| CS 22830 | pD991-AP3 | Ws-2 | ~37,800 | ~408,140 | 11 | 4 <i>aux1</i> , 1 <i>pin2</i> , | | I I |

| | | | | | | | | |
|----------|-----------|------|---------|----------|----|------------------|----------------------|----|
| | | | | | | 1 <i>tir1</i> | <i>crc4(za212)</i> | I |
| | | | | | | | <i>crc2(lb2)</i> | I |
| | | | | | | | <i>ckrc2-2(lb72)</i> | II |
| | | | | | | | <i>ckrc5(lb113)</i> | II |
| CS 6502 | 3850:1003 | Ws-2 | ~6,500 | ~71,580 | 4 | | | |
| CS 84442 | 3850:1003 | Ws-2 | ~4,000 | ~40,050 | 39 | 32 <i>pin2</i> , | <i>crc3(c5)</i> | I |
| | | | | | | 1 <i>aux1</i> , | | |
| Total | | | ~59,600 | ~632,830 | 96 | 64 | 8 | |
