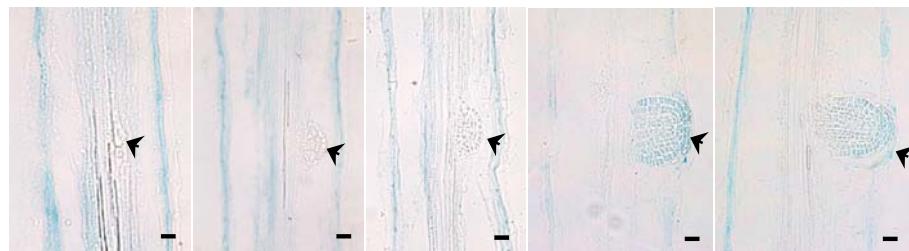
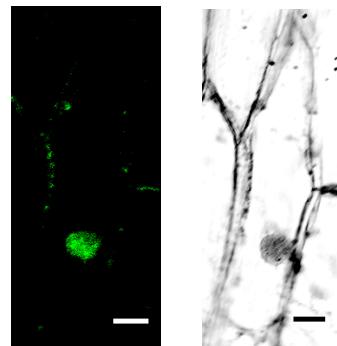


Supplemental data. Zhao et al. (2009). The Wuschel-related homeobox gene *WOX11* is required to activate shoot-borne crown root development in rice.

A



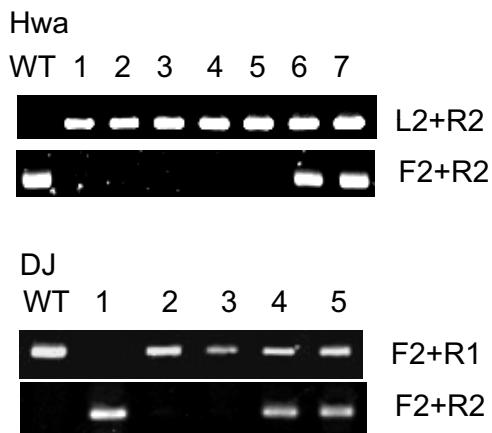
B



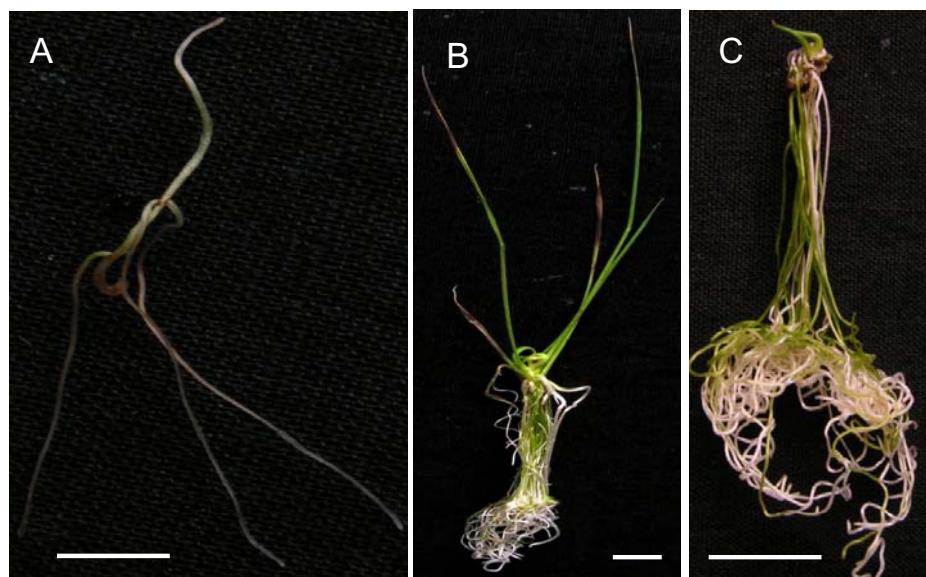
Supplemental Figure 1. Expression of WOX11p-GUS and Nuclear localization of WOX11-GFP protein

A. WOX11p-GUS expression during the course of lateral root initiation. GUS activity was detected after the initiation of the lateral roots . Bars=25 mm.

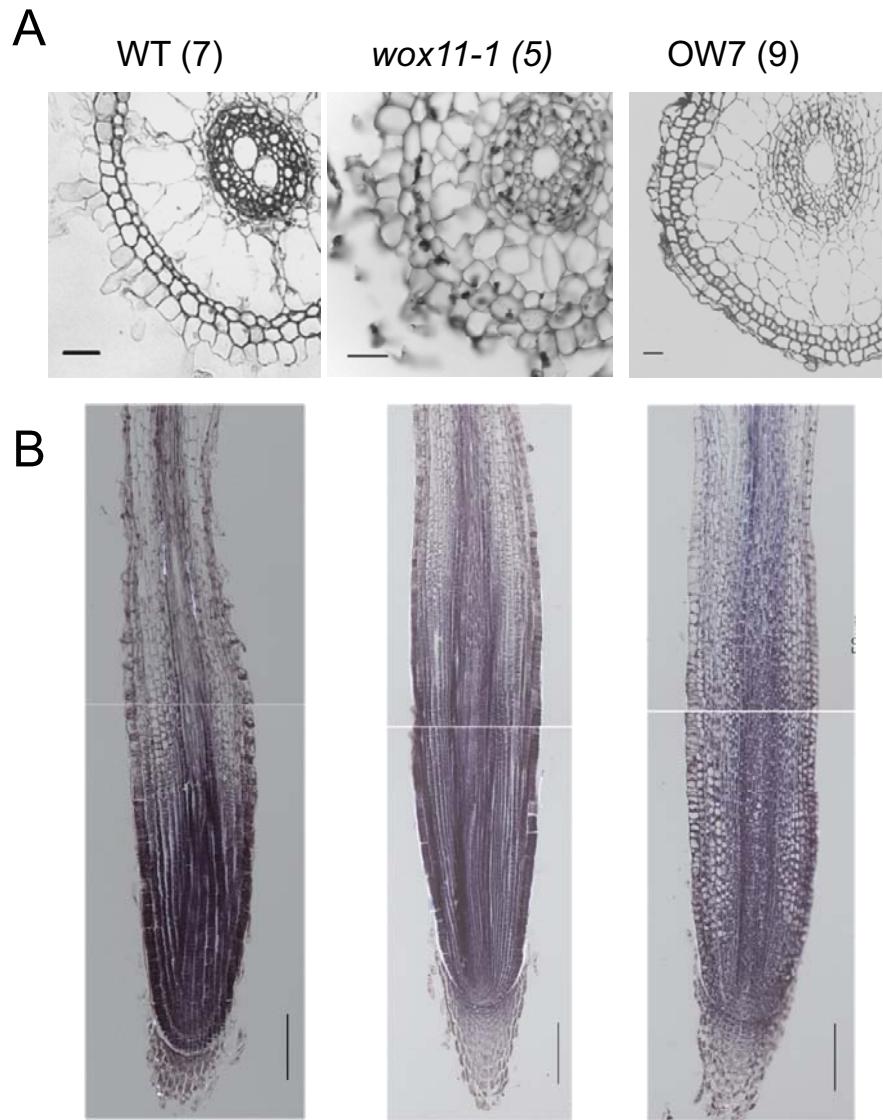
B. Fluorescence image of an onion epidermal cell expressing WOX11-GFP (left) merged with a bright-field image (right) Bar=50 mm.



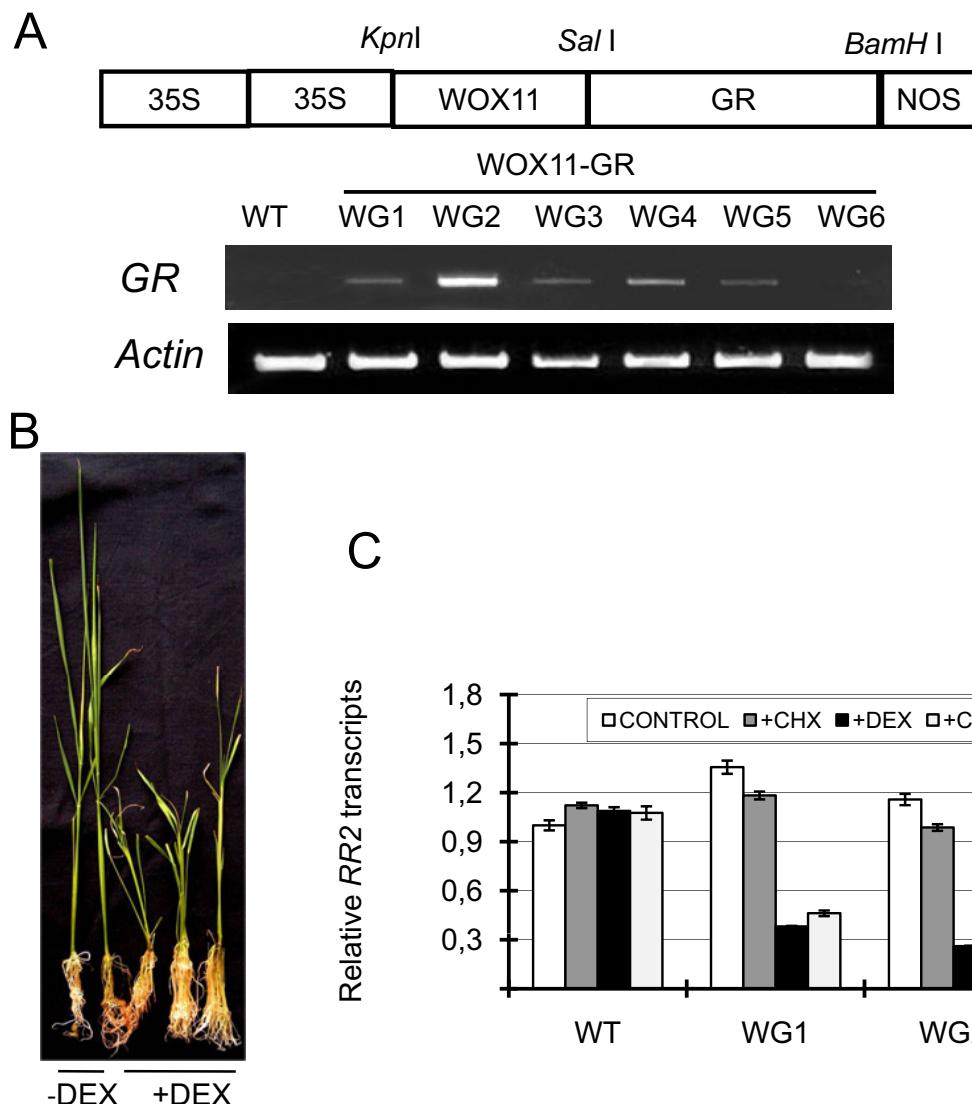
Supplemental Figure 2. Genotyping of the *WOX11* locus in *wox11-1* (upper panel) *wox11-2* (lower panel) and the respective wild type (Hwa and Dongjing). The primer sets used in the PCR tests are indicated on Fig. 2A and included in Supplemental Table 1 .



Supplemental Figure 3. Severe phenotypes found in the T0 population of *WOX11* over-expression plants. A. transgenic plant with the root-like shoot. B and C: transgenic plants with a reduced shoot but an extensively developed root system. Bars=2cm.

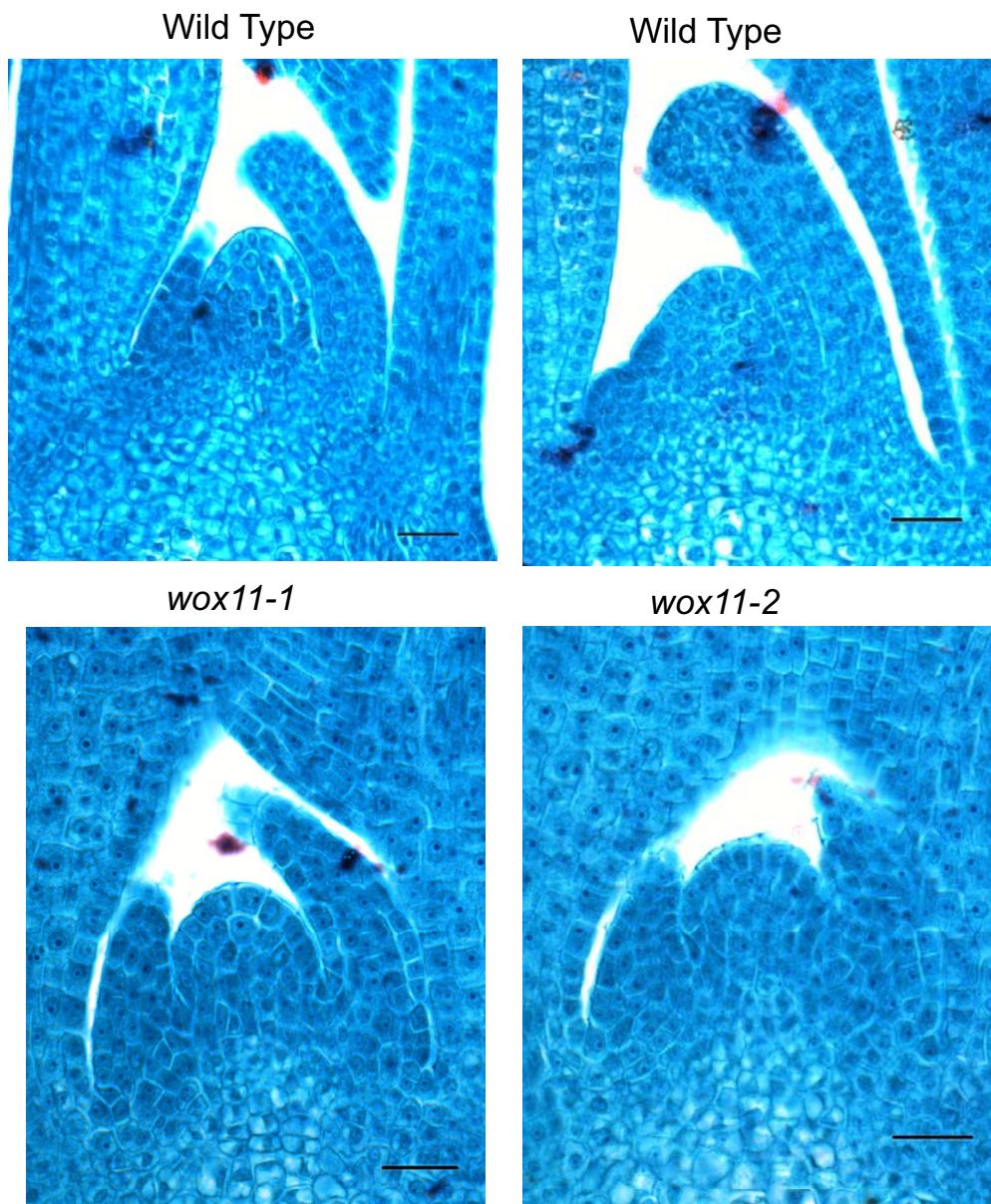


Supplemental Figure 4. Comparison of crown root cross (A) and longitudinal (B) sections of wild type (left), *wox11-1* mutant (middle) and over-expression (OW7) plants (right). Cell layers counted from the cortical and epidermal tissues in A are indicated in parentheses. Bar=25 mm in A, 50 mm in B.



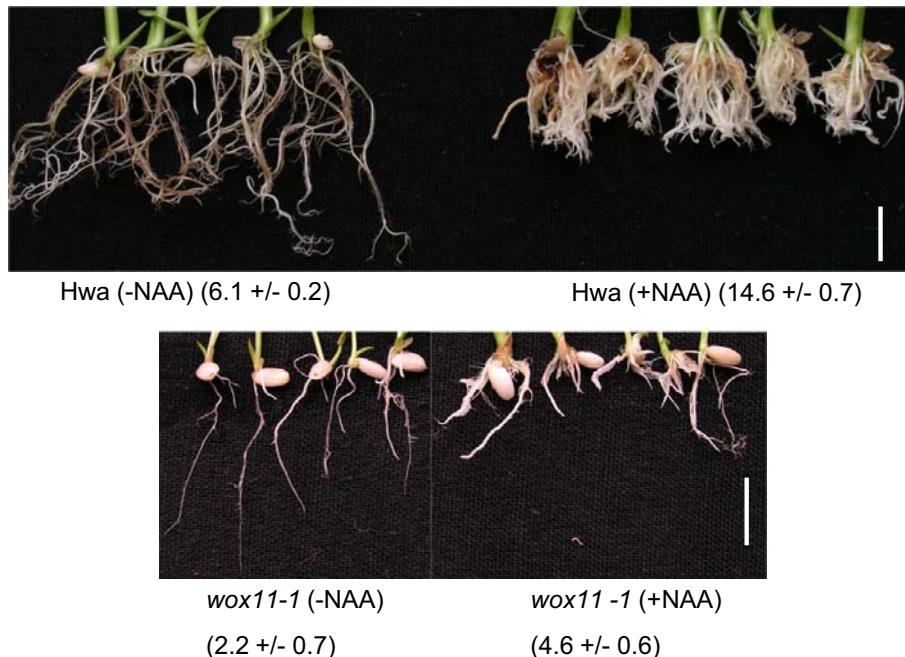
Supplemental Figure 5. Production of transgenic lines expressing the *WOX11-GR* fusion

- Identification of transgenic lines expressing the *WOX11-GR* fusion by RT-PCR. The binary vector containing the translation fusion between *WOX11* and the mouse glucocorticoid receptor (GR) under the control of the double CaMV35S promoter was used to produce transgenic plants. RT-PCR analysis revealed 5 *WOX11-GR*- expressing lines (WG1-5).
- WOX11-GR transgenic plants show extensive root production in the presence of DEX. The DEX was applied every 7 days for 4 times.
- Relative expression levels of *RR2* in wild type and in 3 *WOX11-GR* transgenic plants treated with or without DEX, CHX or DEX plus CHX. The transcript levels were normalized with the *actin* mRNA. The wild type control was assessed as 1. Bars are means +/- SD from 3 biological repeats.

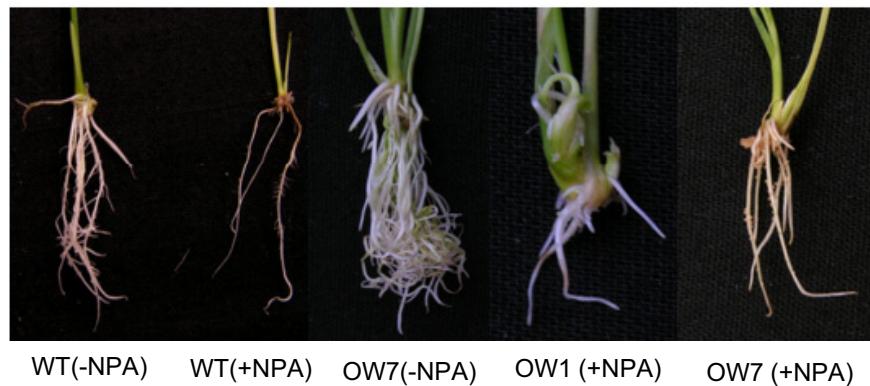


Supplemental Figure 6. Comparison of shoot longitudinal sections from wild type and *wox11* mutants. Shoot apices of 7-day old seedling were stained with hematoxylin, fixed in FAA and sectioned to 8 mm thickness and photographed under a light microscope. Bar = 25 mm.

A



B



Supplemental Figure 7. Effect of auxin and an auxin inhibitor on crown root growth
A. The mutation of *wox11-1* impaired auxin induction of crown root production. Wild type (Hwa) and *wox11-1* were treated with or without NAA. Average root numbers and SD from 20 plants are indicated.

B. Inhibition of ectopic root production by NPA treatment on *WOX11* over-expression plants (OW) and normal crown root growth in wild type (WT). Bars= 1 cm.

Supplemental Table 1: Primers used in the study.

| Experiment | Primer name | Sequence |
|--|----------------------|---|
| wox11 T-DNA mutant characterization | F2 | 5'GGCTGATACTAGCTAGGACACTGA |
| | R2 | 5'CTCAACTCGATCAAGACGACCT |
| | L2 | 5'CTAGAGTCGAGAATTCACTACA |
| | R1 | 5'CCACAGTTTCGCGATCCAGACTG |
| RT-PCR analysis of <i>WOX11</i> expression in <i>wox11</i> mutants | F3 | 5'AACAGCGGCATGGTGAACCC |
| | R3 | 5'GCCACGATGAGGACGACGAG |
| | <i>Actin</i> -F | 5'CTCAACCCCCAAGGCTAACAG |
| | <i>Actin</i> -R | 5'ACCTCAGGGCATCGAAC |
| Vector constructions | <i>OXWOX11</i> -F | 5'GGGGGTACCGCAGCTCTAGGTGTTCGAC |
| | <i>OXWOX11</i> -R | 5'GGGGGATCCATCGACGAATCGCTCAACTC |
| | <i>WOX11gus</i> -F | 5'GGGGTCGACCCCAATCAAATGCTCTGCC |
| | <i>WOX11gus</i> -R | 5'GGGGGATCCCTGCCTGTTGGTGTGCGA |
| | <i>WOX11</i> /RNAi-F | 5'GGGACTAGTGGTACCCGACACCGAACAAAGGC |
| | <i>WOX11</i> /RNAi-R | 5'GGGGAGCTCGGATCCGGAGGAGATCCGAC |
| | <i>WOX11gfp</i> -F | 5'GGGGGTACCGACACCGAACAAAGGCAGCTA |
| | <i>WOX11gfp</i> -R | 5'GGGGGATCCAGACGACCTCGTGACCAGG |
| | <i>WOX11gr</i> -F | 5'GGGGGTACCGACACCGAACAAAGGCAGCTA |
| | <i>WOX11gr</i> -R | 5'GGGGTCGACAGACGACCTCGTGACCAGG |
| Realtime PCR | <i>GR</i> -F | 5'GTCGACAGATCCTGAAGCTCGAAAAAC |
| | <i>GR</i> -R | 5'GGATCCACCGGAAACAGGATTCAATG |
| | <i>WOX1132a</i> -F | 5'ACAGGTACCGACGACGACAAGATGGACGGCGGCCACAGC |
| | <i>WOX1132a</i> -R | 5'ACAGGATCCTCAAGACGACCTCGTGACCAGG |
| | <i>RR1</i> -F | 5'AGGATCAGCAGATGCATGAATG |
| | <i>RR1</i> -R | 5'GAGACGCTGTACGTCCCTGCTT |
| | <i>RR2</i> -F | 5'ACGATCTCTCAAAGCCATCAAG |
| | <i>RR2</i> -R | 5'TGAGAGGCTTAAGGATGAAATCCT |
| | <i>RR3</i> -F | 5'AGGGTTCGATCTCCTCAAGAG |
| | <i>RR3</i> -R | 5'GAATTCTCCGACGACATTAGC |
| | <i>RR5</i> -F | 5'ACCGAATGTGAGCATGATTATCA |
| | <i>RR5</i> -R | 5'CCTTGACCTTCTCAGGAGTTCTA |
| | <i>RR6</i> -F | 5'GTCCCCAACGTCAACATGATC |
| | <i>RR6</i> -R | 5'CACGTTCTCCGACGACATGAT |
| | <i>RR7</i> -F | 5'TGCTCAAGAAGATCAAGGAATCG |
| | <i>RR7</i> -R | 5'GGCACGTTCTGACGACATTAT |
| | <i>IAA5</i> -F | 5'GAAATTGAAAATCATGAGAGGATCTG |

| | | |
|------------------------------|---------------------|-------------------------------------|
| | <i>IAA5</i> -R | 5'TGCTCTGCCCTGACTGCTCTA |
| | <i>IAA11</i> -F | 5'CGACGTGCCATGTACAAGA |
| | <i>IAA11</i> -R | 5'TGGTGAAGGAGGTGAACATGTT |
| | <i>IAA23</i> -F | 5'TGCCCACCTACGAGGACAAG |
| | <i>IAA23</i> -R | 5'TTGCAGGACTCGACGAACATC |
| | <i>IAA31</i> -F | 5'CGACGTCCCATTGAGATGT |
| | <i>IAA31</i> -R | 5'TTGCTCCTAGGCCTTTGCTT |
| | <i>WOX11</i> -F(rq) | 5'CCAGATGGCGAGAGCTACT |
| | <i>WOX11</i> -R(rq) | 5'CGTGCCATCGATCAATCAA |
| | <i>Actin</i> -F(rq) | 5'TGTATGCCAGTGGTCGTACCA |
| | <i>Actin</i> -R(rq) | 5'CCAGCAAGGTCGAGACGAA |
| <i>In situ</i> hybridization | <i>RR2situ</i> -F | 5'GGGACTAGTGGTACCGTCGTCGGAGAACATGCC |
| | <i>RR2situ</i> -R | 5'GGGGAGCTCGGATCCGACCATCTGTGCAGGAGC |
| Gel shift assays | S1-F | 5'GACCCCCGGCGATTAAATGGTTAATTAA |
| | S1-R | 5'TAATTAACCATTAAATGCCGGGGGT |
| | S2-F | 5'GACCCCCGGCGATCCGGGTTAATTAA |
| | S2-R | 5'TTAATTAACCCGGGATCGCCGGGGGT |
| ChIP | P1-F | 5' CGCCTTAGGCTTCATCAATC |
| | P1-R | 5'TATTGGCATCGACGTGGTTA |
| | P2-F | 5'CGGATAGTTGTCTGAATTG |
| | P2-R | 5'GAATTCAACTGTGCGATG |
| | P3-F | 5'CATCGCACAGTTGAATT |
| | P3-R | 5'CGTGAGGTAGTACACACGG |
| | P4-F | 5'GCCAATACAATAACCAT |
| | P4-R | 5'GAGGGAAATTCAAGATA |