

A green cotton boll is the central focus of the image. Overlaid on the boll is a world map, with the landmasses of North and South America clearly visible. The boll is set against a solid yellow background.

2012 ICGI research conference

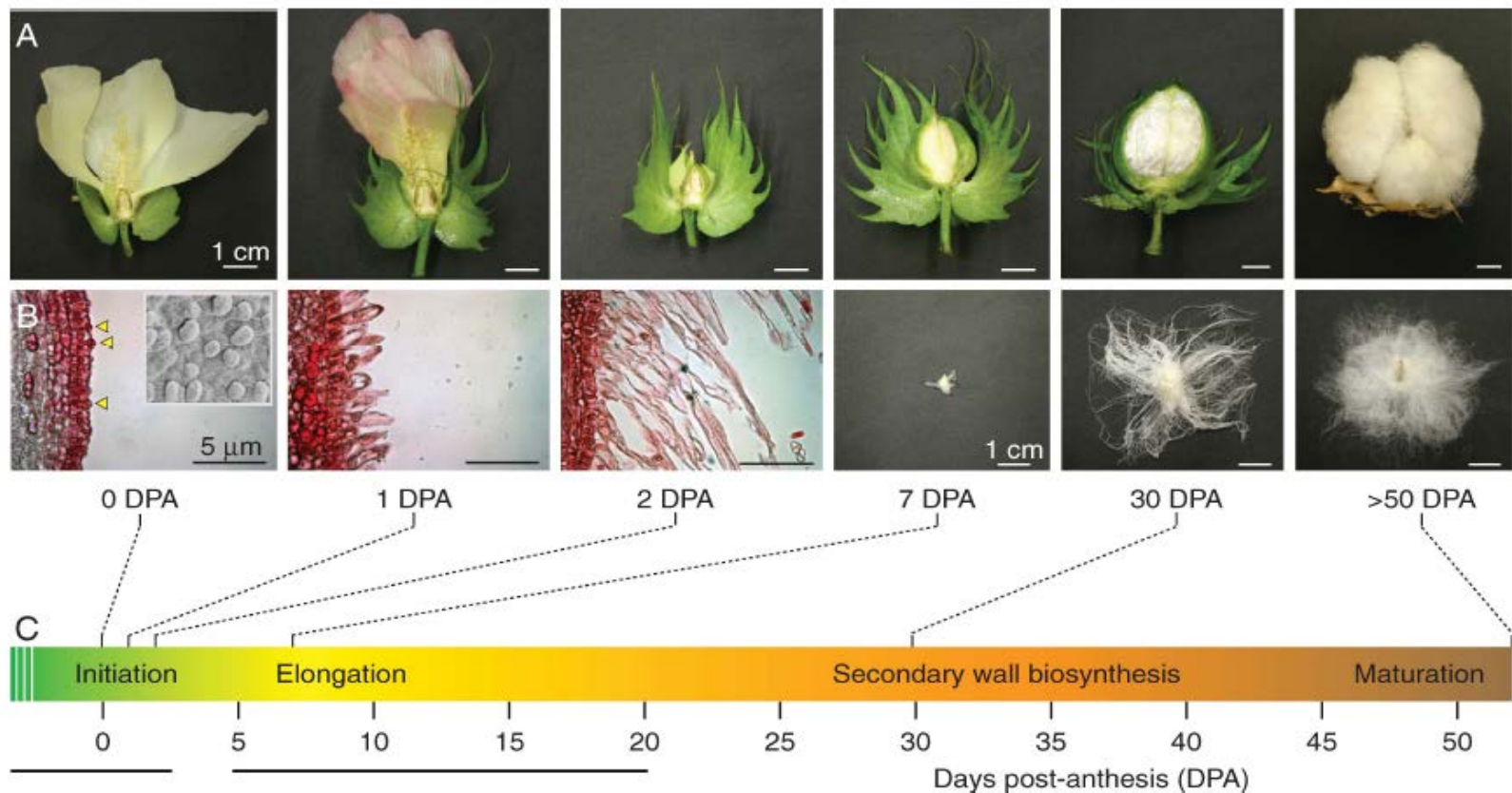
Increased endogenous brassinosteroid levels promote elongation of cotton fiber

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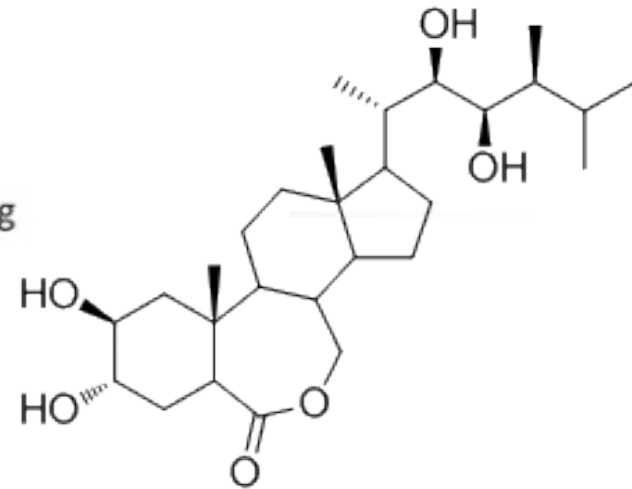
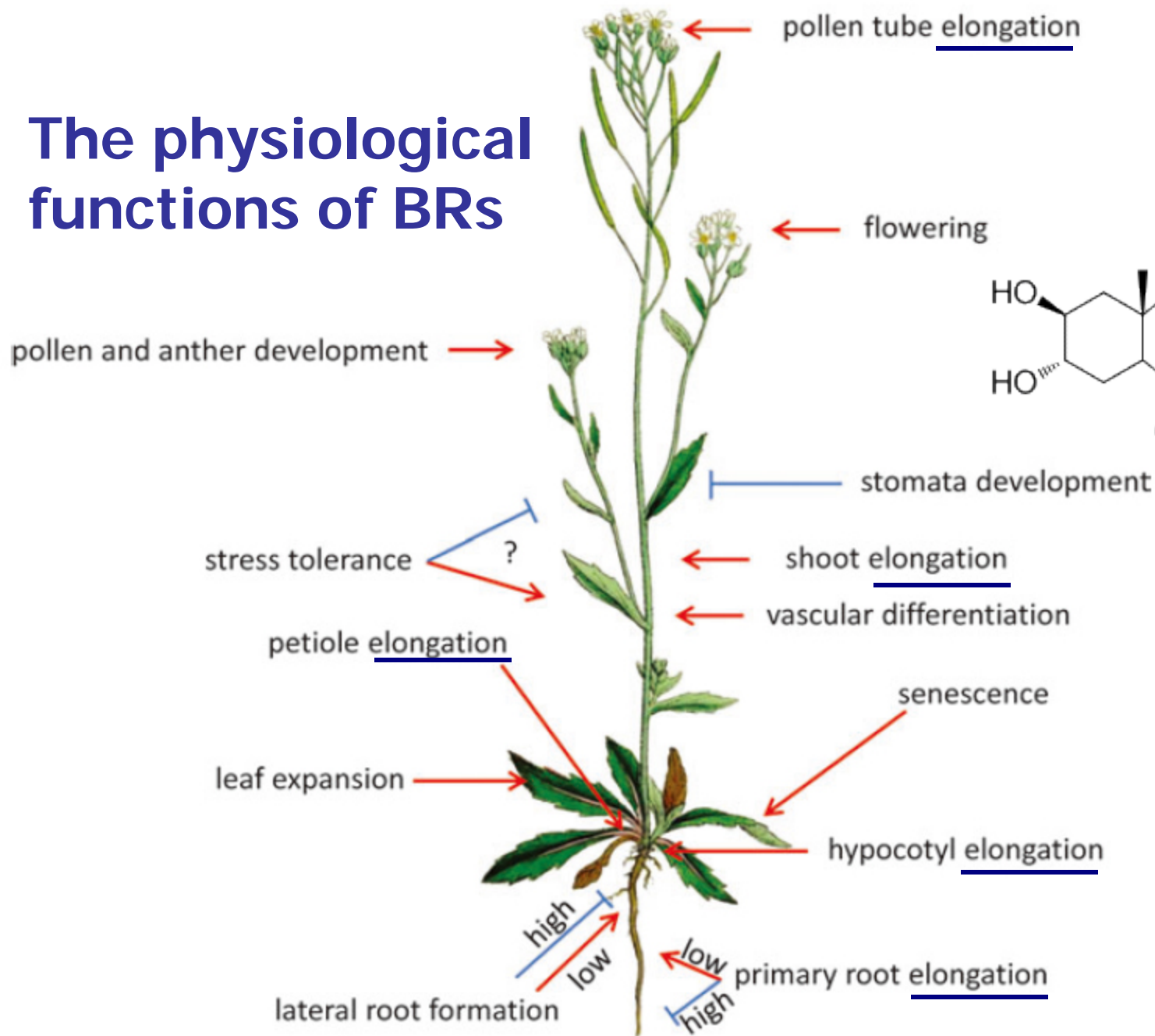
Background

The growth and development of cotton fibers



Cotton fiber developmental stages (Lee *et al.*, 2007).

The physiological functions of BRs





BRs regulate cotton fiber growth and development

BR treatment promotes cotton fiber elongation in ovule culture or plant

BR biosynthetic and signaling genes highly expressed in fast elongating fibers

BRs accumulated in fiber cells

Physiology



Molecular biology



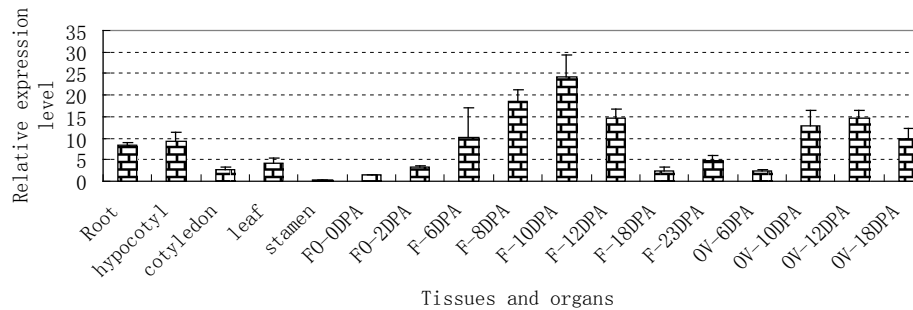
Biochemistry



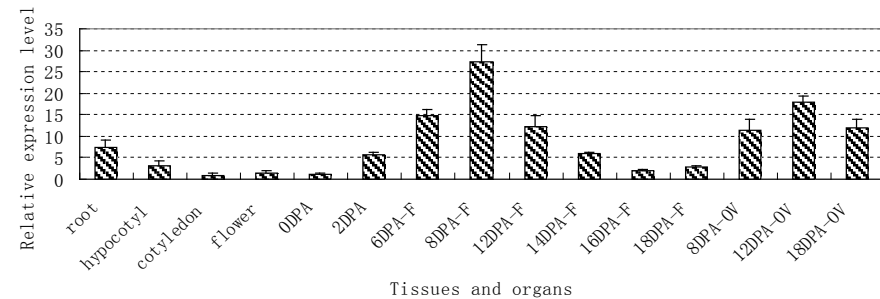
Exogenously applied brassinosteroids (BRs) advance cotton fiber yield and quality in agricultural industry

- **United States Patent 5880110 :**
Production of cotton fibers with improved fiber characteristics by treatment with brassinosteroids
- **Synthesis and practical applications of brassinosteroid analogs**
By S. Hayat and A. Ahmad, Brassinosteroids: Bioactivity and Crop Productivity (2004), ISBN 978-1-4020-1710-0
“...foliar sprays of 28-homobrassinolide significantly increased wheat, riceand cotton seed yield.”
- “Yunda-120”, a BR commercial preparation for cotton production in China.

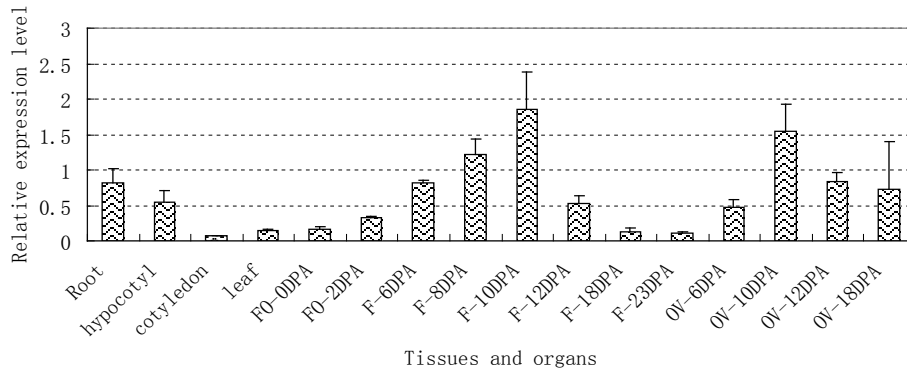
The genes involved in biosynthesis preferentially expressed in fiber cell elongation stage



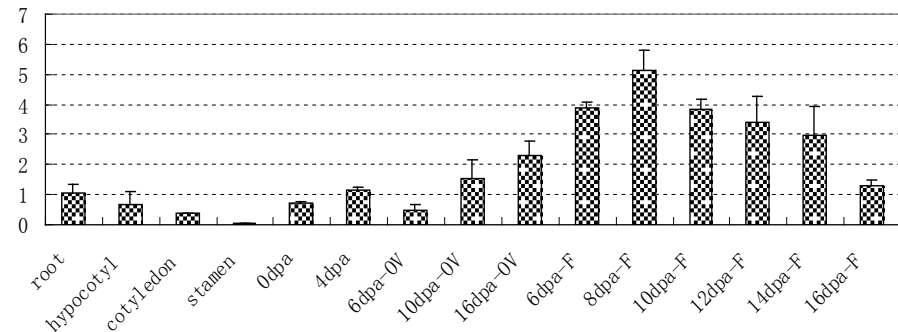
GhSMT2



GhDWF5



GhDWF1

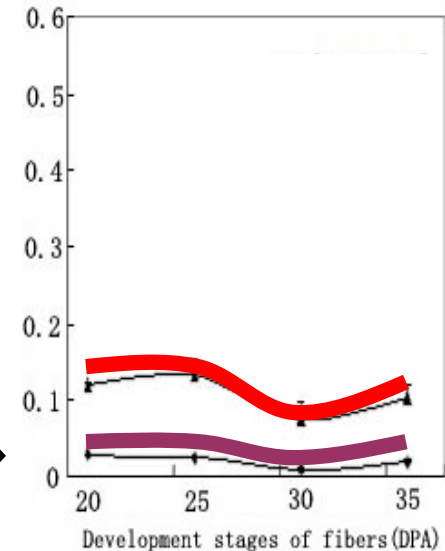
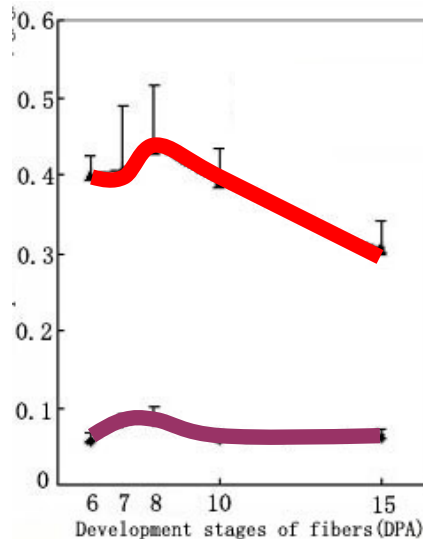
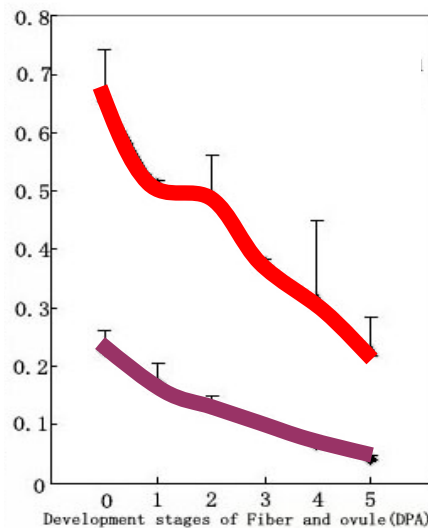


GhHYD1

The content of phytosterols, intermediates in up-stream pathway of BR biosynthesis is higher in fiber cell development

— sitosterol

— campesterol



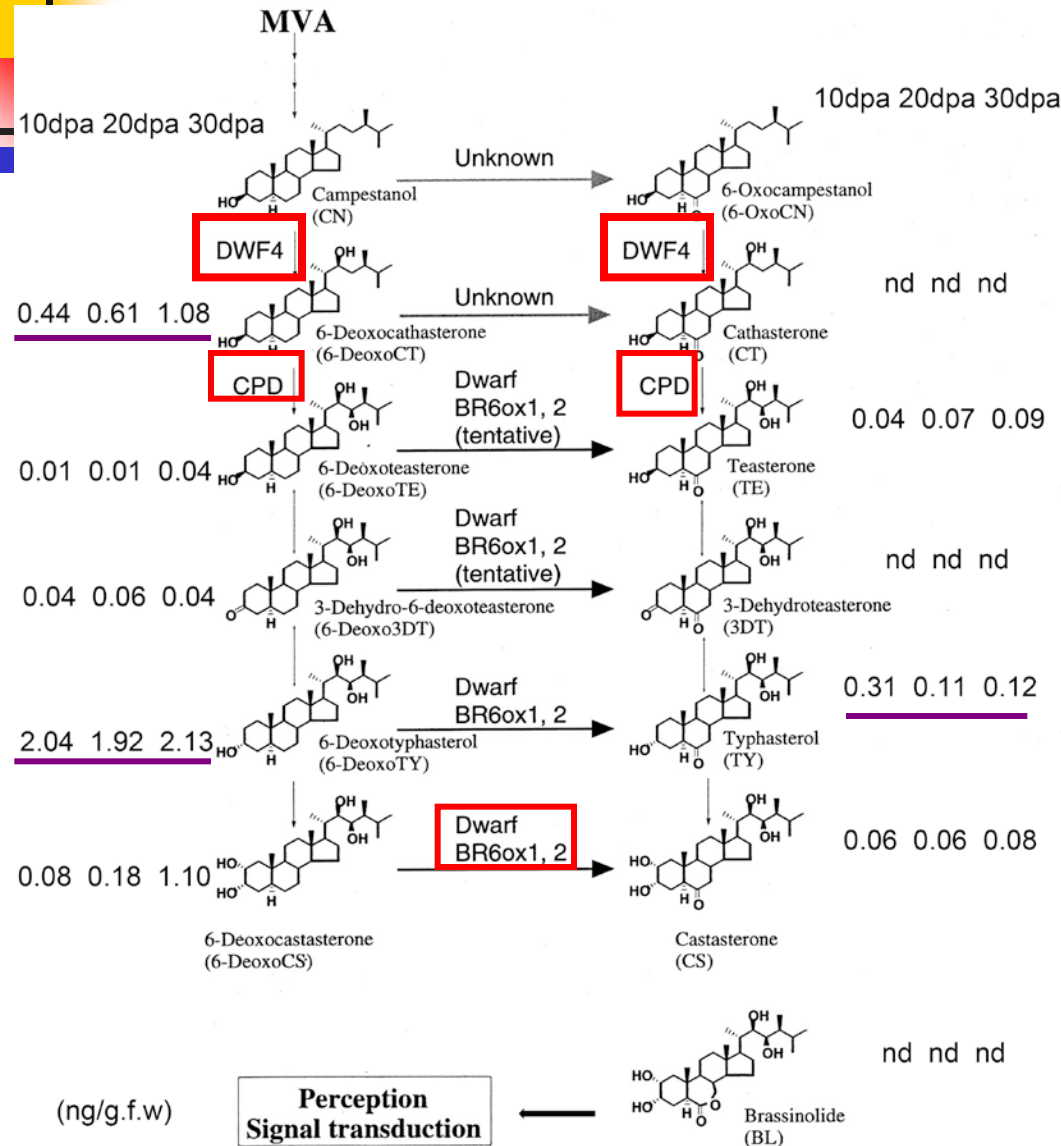
Fiber cell initiation

elongation

Fiber rapid elongation

Secondary cell wall deposited

The intermediates in down-stream of BRs biosynthesis pathway



1: the late-C pathway is a main pathway;

2: the content of 6-DeoxoCT、6DeoxoTY and TY are higher than other.



What will happen when the endogenous BRs increased

What is the molecular mechanism of BRs on regulating fiber elongation

We have selected the GhDWF4 gene

DWF4 gene is tightly controlled to express in tissues

Bioactive BRs accumulated in the parts with highly expressed *DWF4*

DWF4 catalyzes the C₂₂ α -hydroxylation in BR biosynthetic pathway

DWF4 is a critical BR biosynthetic gene, catalyzing a flux-determining step

***DWF4* gene was closely related to bioactive BRs**



Azpiroz, R., Wu, Y., LoCascio, J.C., and Feldmann, K.A. (1998). An *Arabidopsis* brassinosteroid-dependent mutant is blocked in cell elongation. *Plant Cell* 10: [219-230](#);

Choe, S., Dilkes, B.P., Fujioka, S., Takatsuto, S., Sakurai, A., and Feldmann, K.A. (1998).

The *DWF4* gene of *Arabidopsis* encodes a cytochrome P450 that mediates multiple 22 α -hydroxylation steps in brassinosteroid biosynthesis. *Plant Cell* 10: [231-244](#).

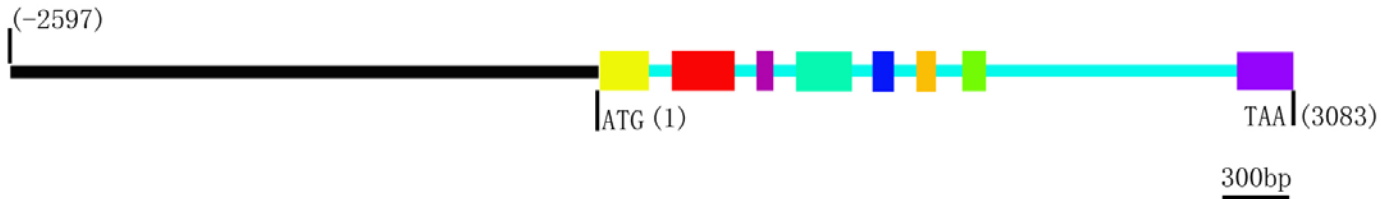
Cloning of *GhDWF4* gene

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AtDWF4 MFETEHHTL-LPLLPSILSLFLILKRRNK-----TFENLPPGKSGWPELGETIGYLPYATTLGDF
GhDWF4 MPDSEP---T-ELLPSILSLFLILKRRNK-----RYNLPPGNMGWPELGETIGYLRFPYSATSVGEF
OsDWF4 MASITS---ELFFFFFIILALLTFYTTTAKCHGGHWRGGTTPAKRKEMNLPPGAAGWPELGETIGYLRFPYSATSVGEF
ZeDWF4 MCSITLNMCDLDFILASGLVLAFLILKLVKRRIT-----NNGSTRNLPPGNMGWPELGETIGYLRFPYSATTIGKF

                ANCHOR REGION                                PROLINE

AtDWF4 MQQHVSKYGIYRSNLFGEPTIVSADAGLNRFILQNEGRLFECYPRSIGGILGKWSMLVLVGDHHRMRSISLNFLSHAR
GhDWF4 MQQHISRYGNIYKSNLFGEPTIVSADAGLNKFIQNEGRLFECYPRSIGGILGKWSMLVLVGDHHRMRIISLNFLSNAR
OsDWF4 MQQHIARYGKIYRSNLFGEPTIVSADAGLNRYILQNEGRLFECYPRSIGGILGKWSMLVLVGDHHRMRAISLNFLSVR
ZeDWF4 MQQHISKYGIYKSNLFGEPTIVSADAGLNKYILQNEGRLFECYPRSIGGILGKWSMLVLVGDHHRMRQISLNFLSNAR
  
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The genomic structure of GhDWF4 gene

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ZeDWF4 KALKSRVILKLTETKMEPRIR-MDQG-----NGQG-----KLDNDLLEQSSKNSNLTRREQITDVLVLSLLEF

AtDWF4 AGHETSSVITLAIYFLPCCPLAIQQLREEHLEVAFAHQSGETELNDDYKMEFTQCVINETLRIGNVVRFLEHAKIKD
GhDWF4 AGHETSSVITLAIYFLPCCPLAIQQLREEHLEVAFAHQSGETELNDDYKMEFTQCVINETLRIGNVVRFLEHAKIKD
OsDWF4 AGHETSSMIALAIFFLEGCPRAVDELREEHIGIARQRLRGECKLSMEDYKEMVFTQCVINETLRIGNVVRFLEHAKIKD
ZeDWF4 AGHETSSVITSLAVYFLACPTAVRQLREEHEHIVMKKLLGEKYLTDYKMEFTQCVINETLRIGNVVRFLEHAKIKD

Domain A                                Domain B

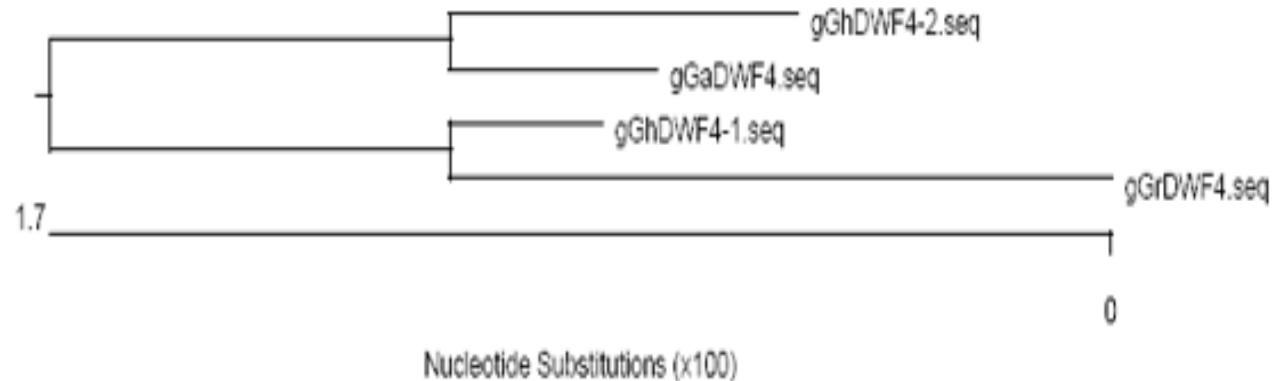
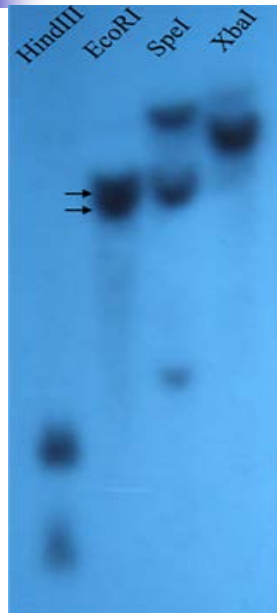
AtDWF4 VHYKGYDIPSGWKVLPVI SAVHLDNRYDQNFENPWRWQQQNGGASSSGGSFTWG-NNYMPFGGGRRLCAGSELAKLE
GhDWF4 IRYKGYDIPCGWKVLPVI SAVHLDPCLFHPQLFNPWRWQQ-NNGSRGASTATSSASSSNYFMPFGGGRRLCAGTELAKLE
OsDWF4 VHYKGYDIPSGWKI LPVL SAVHLDSILYEDPQRFNPWRWKS-----SGSSGGLAQS-SSFMPYGGGTRLCAGSELAKLE
ZeDWF4 VHYKGYDIPCGWKVLPVI SAVHLDPHFHFCPPYTFDPWRWN----ASVTSSTCSIPPSASNFMPFGGGRRLCTGSELAKLE

                Domain C                                HEME-BINDING

AtDWF4 MAVFIHHLVLKFWELVEDDQEFNFPFVDFPGLPIRVSRIL-----
GhDWF4 MAVFIHHLVLNFWELADTDEAFNFPFVDFPGLPIRVFAS-----
OsDWF4 MAVFLHHLVLNFWELVEDDQAFVFPFVDFPGLPIRVHRIAQDDEQE
ZeDWF4 MAIFIHHLVLKFWELVDSDEFNFPYLDFFPKGLPIKIRHRKQSC---
  
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Predicted *GhDWF4* protein is homologous to other DWF4 proteins

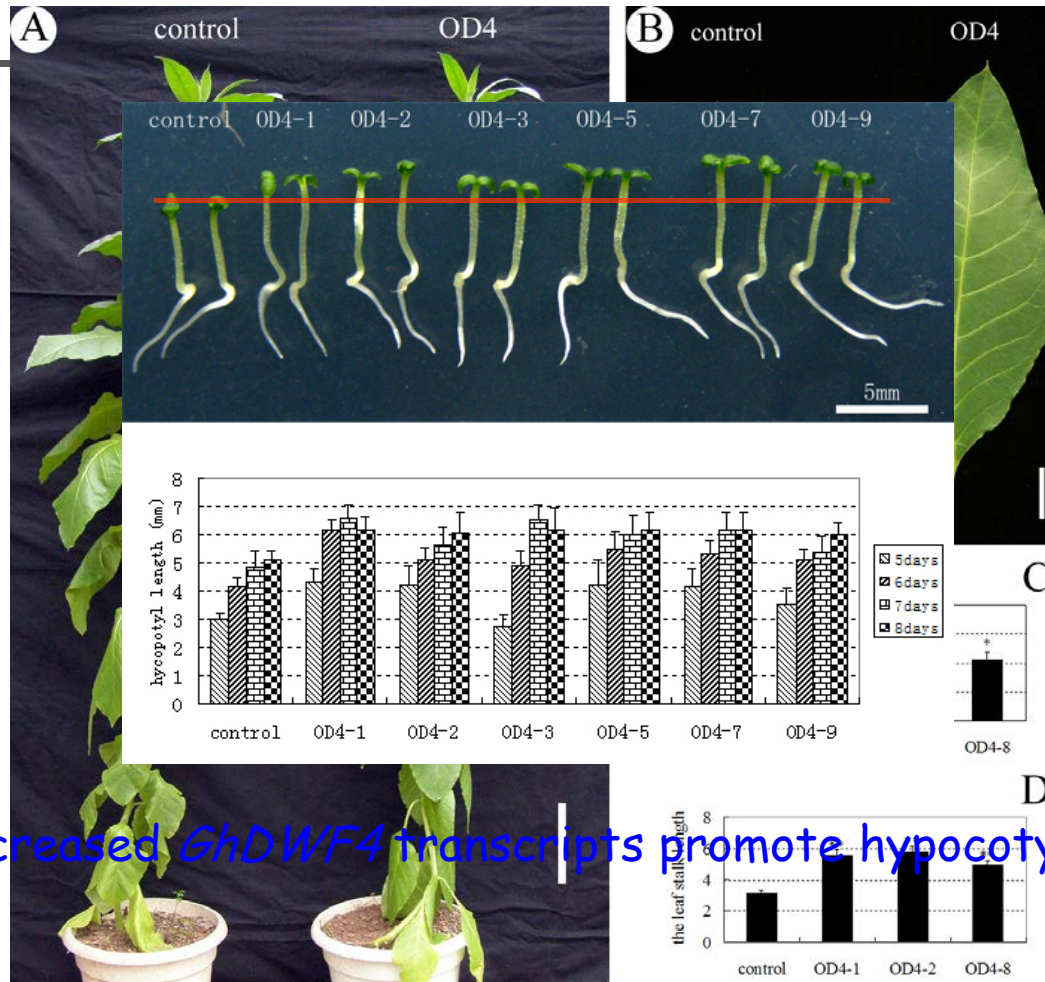
Genetic analysis of *GhDWF4*



The two *GhDWF4* copies are closer to *DWF4* gene from *Gossypium arboreum* (A subgenome) and *Gossypium raimondii* (D subgenome), respectively.

Southern blot of
GhDWF4 gene in
Gossypium hirsutum
(A and D subgenome).

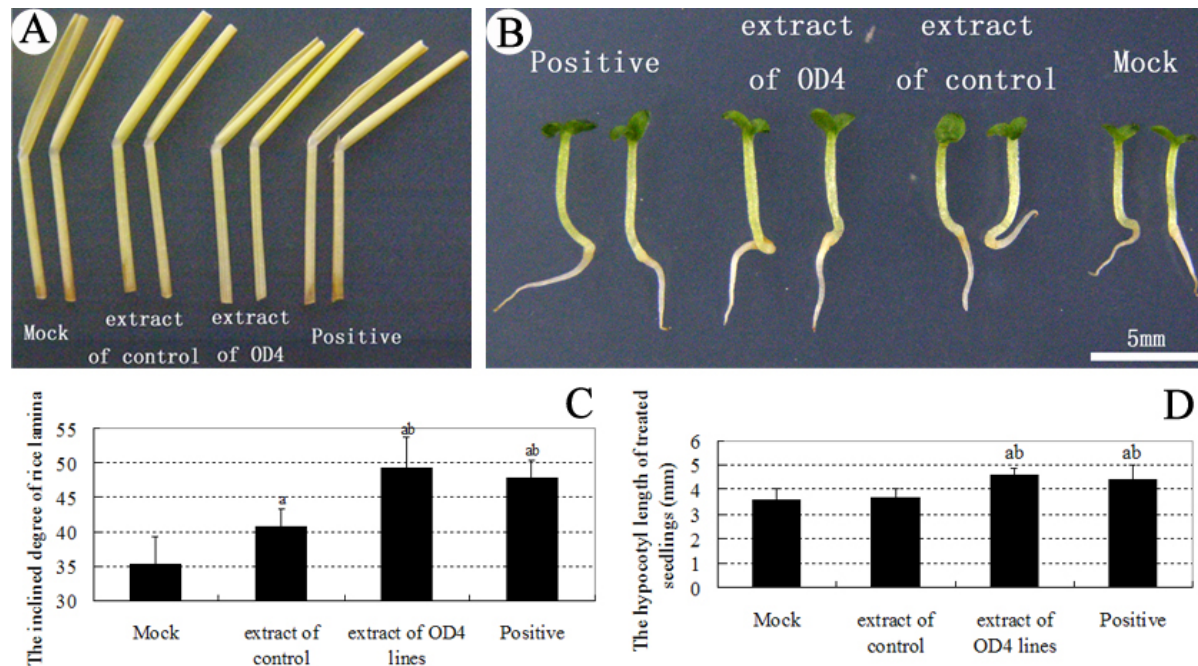
Overexpression of *GhDWF4* regulates tobacco growth



Increased *GhDWF4* transcripts promote hypocotyl elongation.

Overexpression of *GhDWF4* changes leaf shape of tobacco.

Overexpressing *GhDWF4* elevates the bioactive BR levels in transgenic tobacco

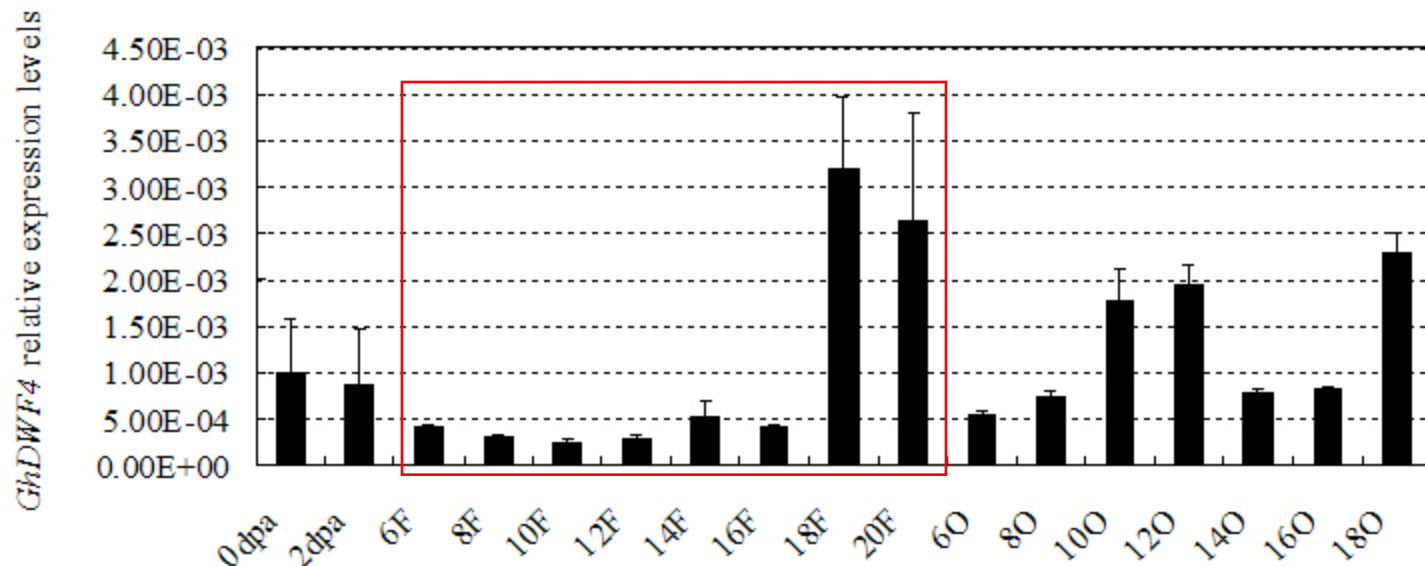


Bioassay analysis of BR extracts.

A and C) rice lamina bending assay.

B and D) germinated tobacco seeds treated by BR extracts.

Quantitative real-time analysis of *GhDWF4* in various tissues and organs of *Gossypium hirsutum*



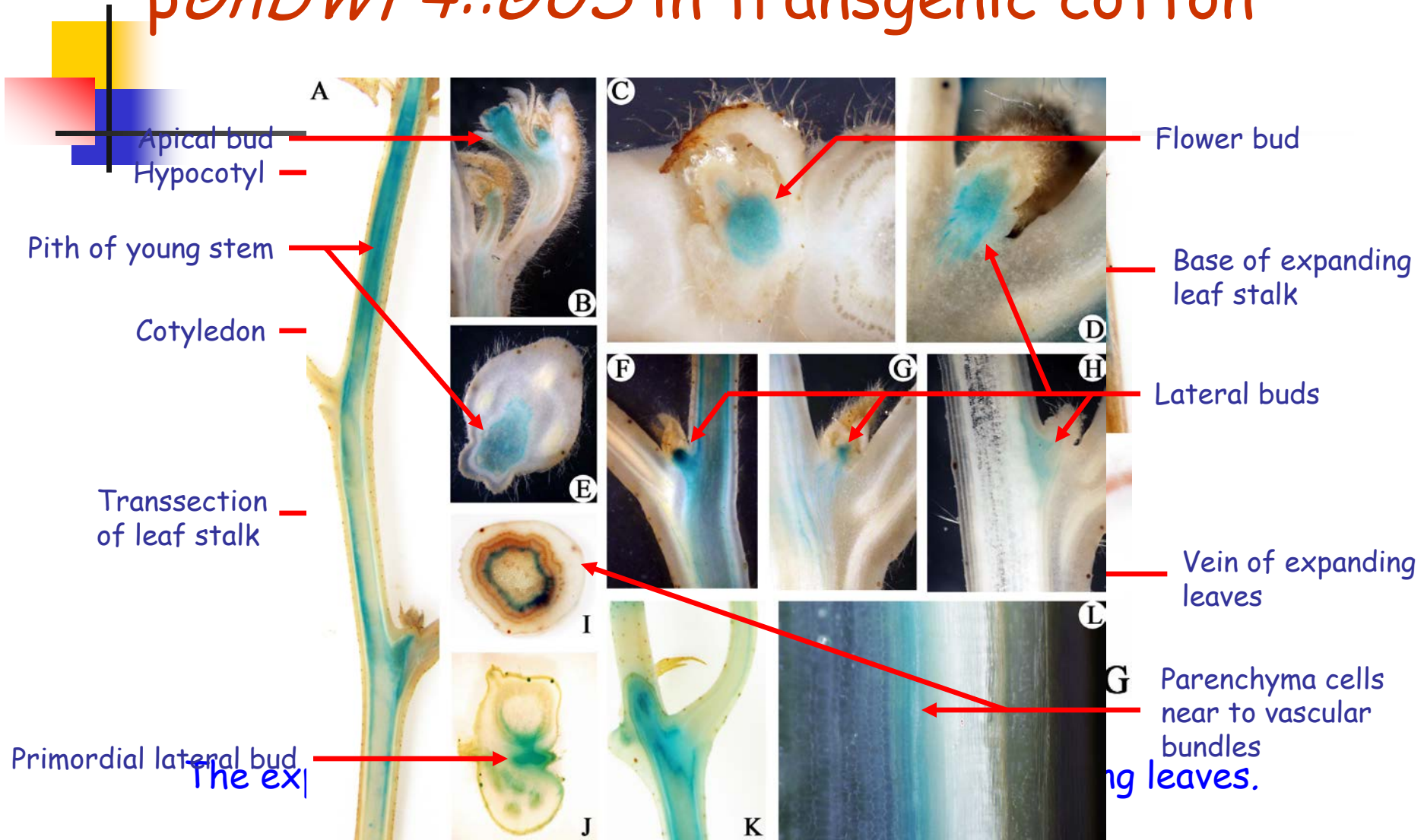
Relative expression levels of *GhDWF4* in various tissues and organs of *Gossypium hirsutum* developing ovules and fibers.

[illegible]

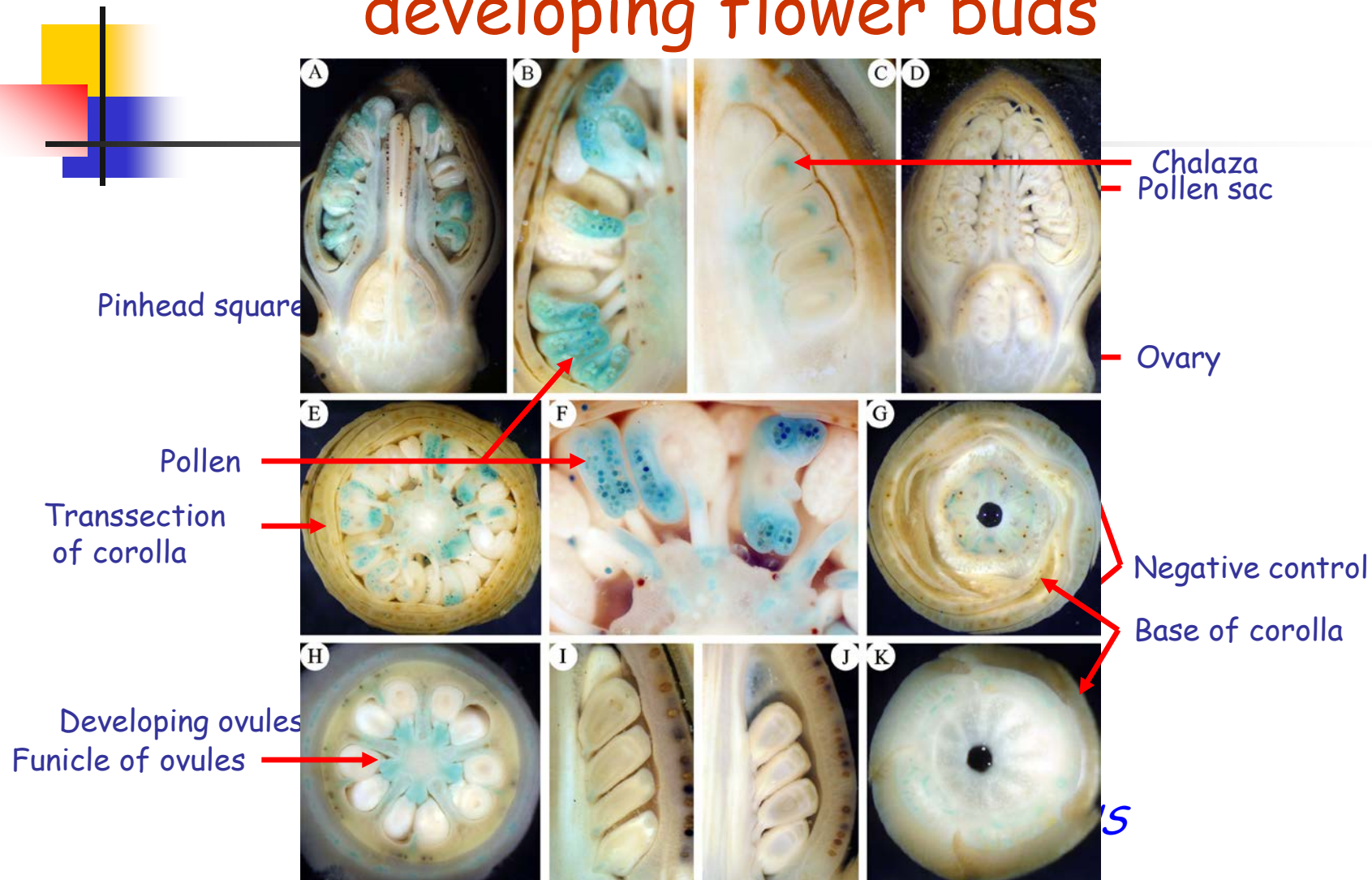
CTGGCATTTTTTTTTCTTTTAGGTAAATAATTCCTATAAAAAAGAAAAGAAAAGAAAAGAAAAGGGGCTATCAGTCTG
pGhDWF4 is more homologous to DWF4 subgenome.

ATG

The spatial and temporal expression of *pGhDWF4::GUS* in transgenic cotton

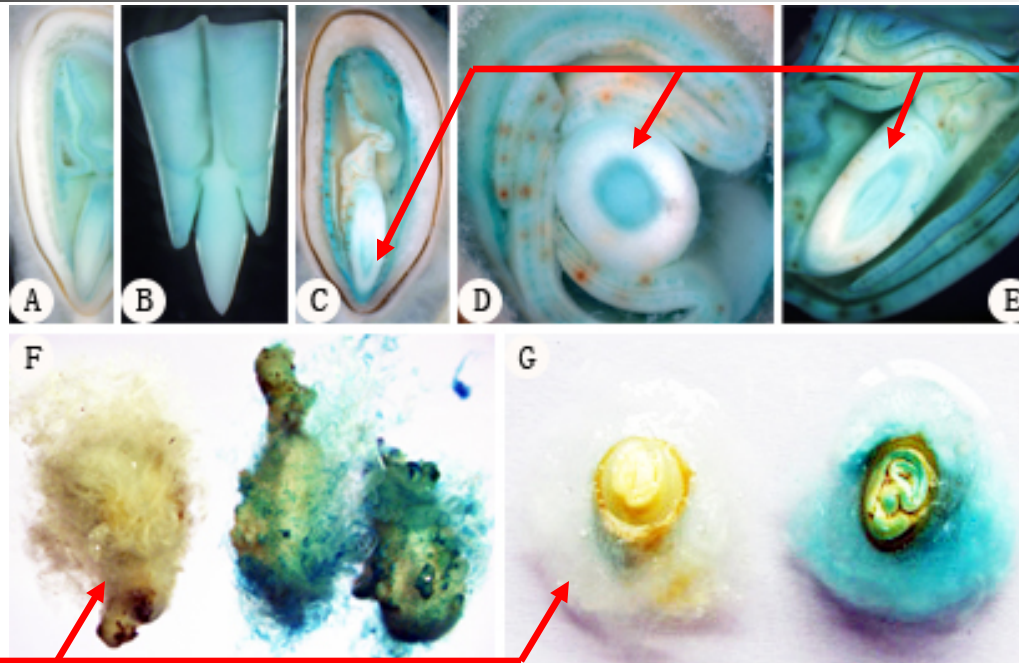


p*GhDWF4*::*GUS* expressed in developing flower buds



The expression pattern of p*GhDWF4*::*GUS* in developing flowers

The expression pattern of *pGhDWF4::GUS* in developing seeds and cotton fibers

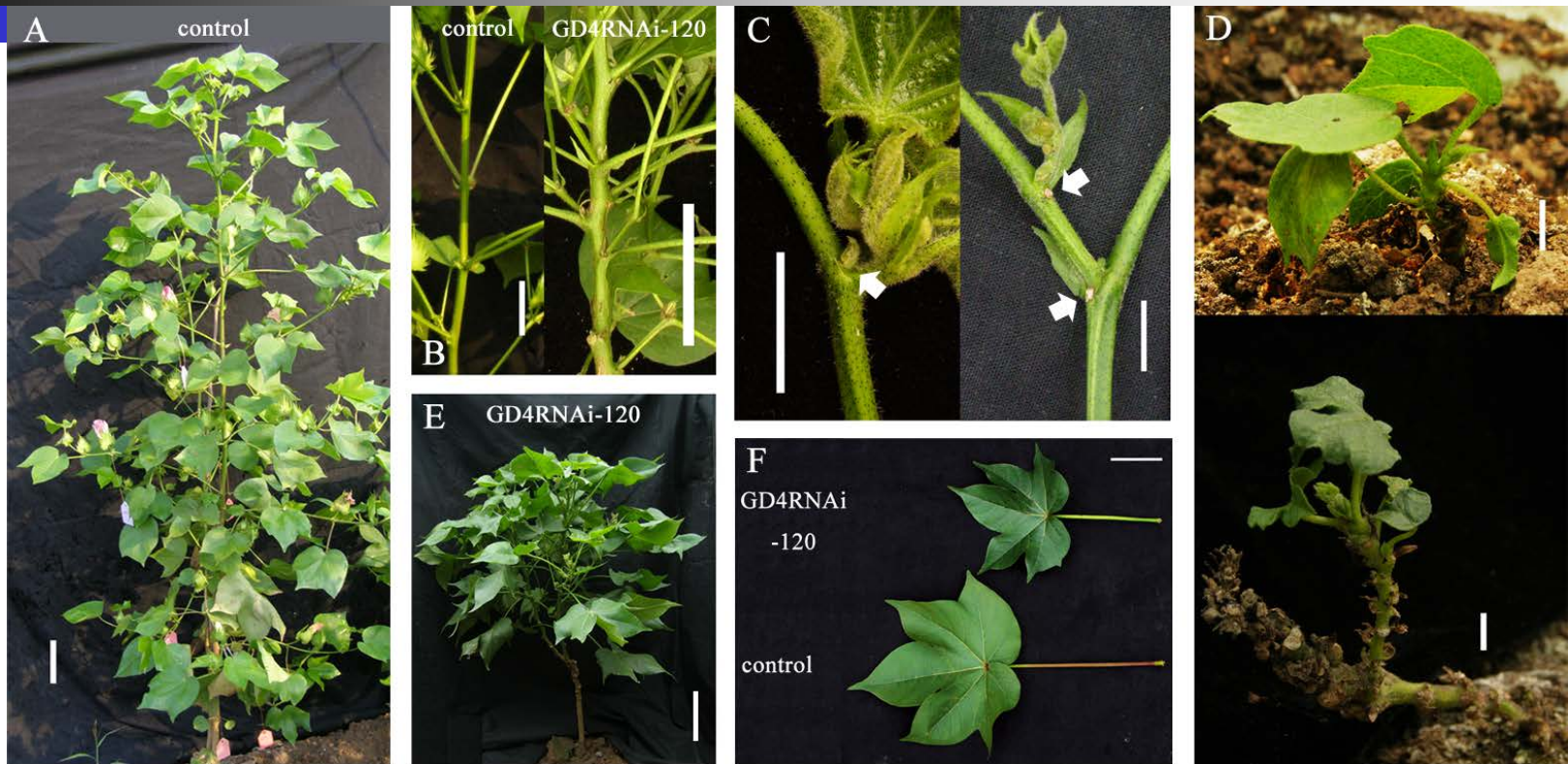


Vascular tissues
of radicle

Negative control

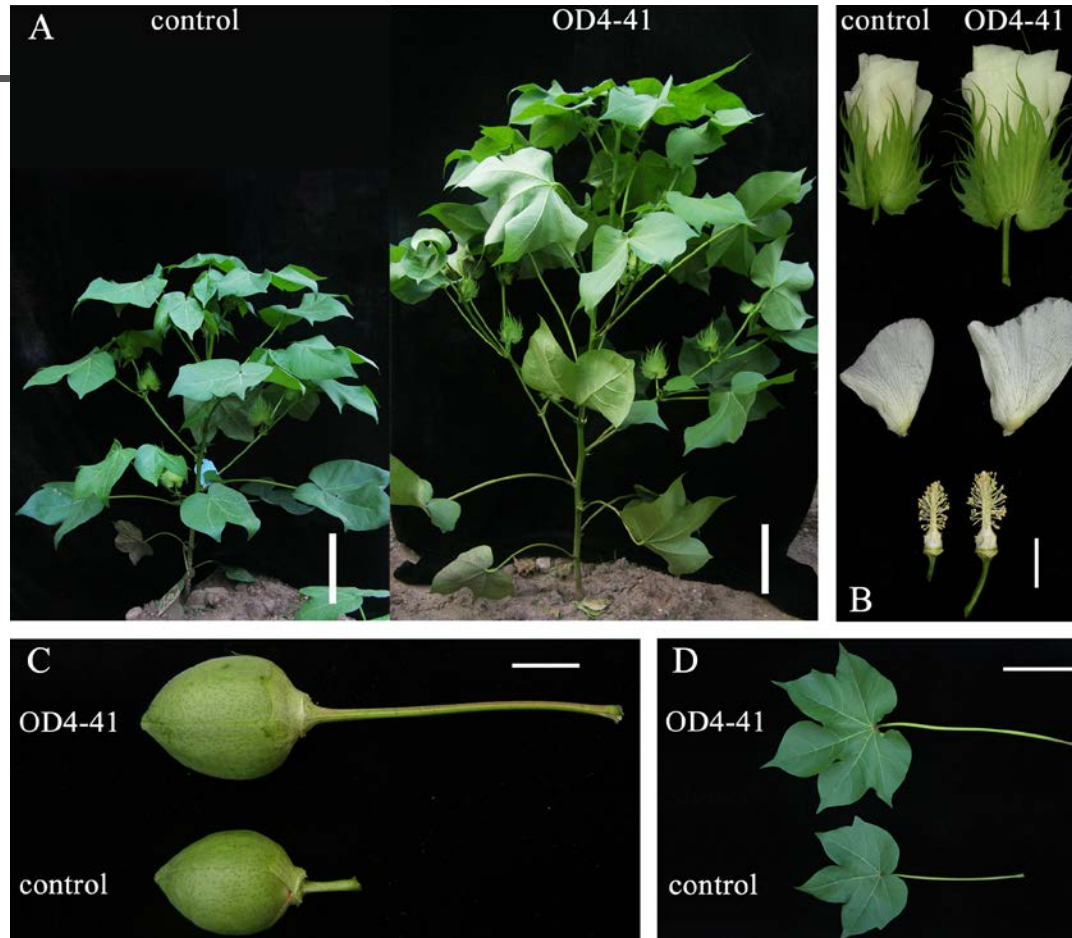
pGhDWF4::GUS expressed in the whole embryos, especially in the vascular tissues of radicle. GUS staining was also detected in cultured fibers *in vitro* and 35dpa fibers collected straightly from *pGhDWF4::GUS* plants.

Repressing *GhDWF4* expression leads to dwarf phenotype



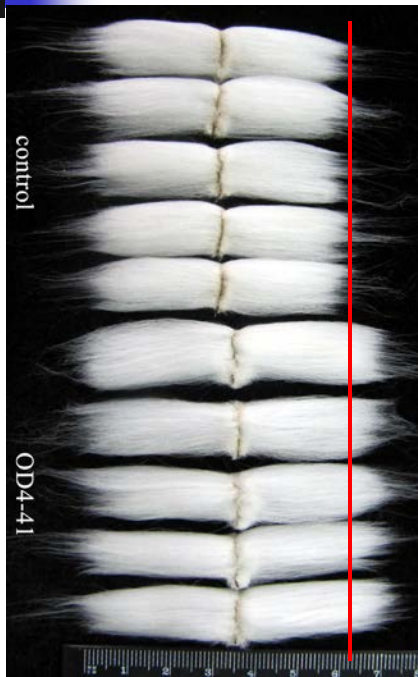
GhDWF4-repressed lines displayed short leaf stalks and internodes, small leaf size, slow growth, and sterile.

Overexpression of *GhDWF4* promotes cotton plant growth

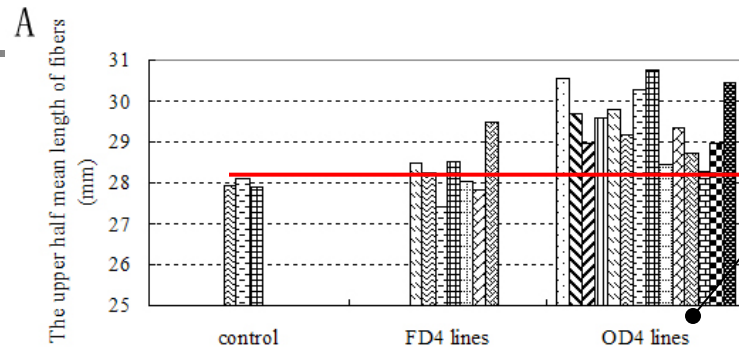


Transgenic cotton plants displayed bigger flowers, bigger bolls, and larger leaf size than control.

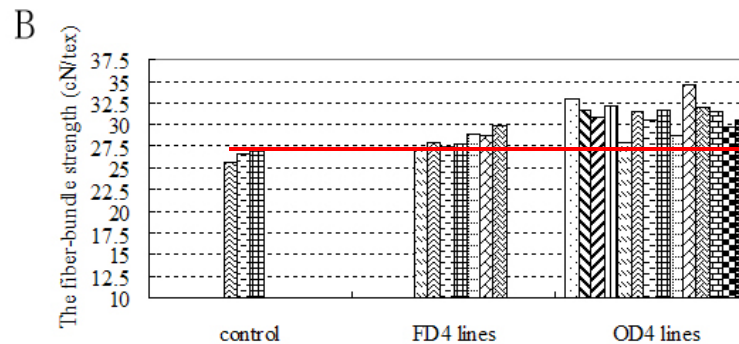
Overexpressing *GhDWF4* improved on cotton fiber quality



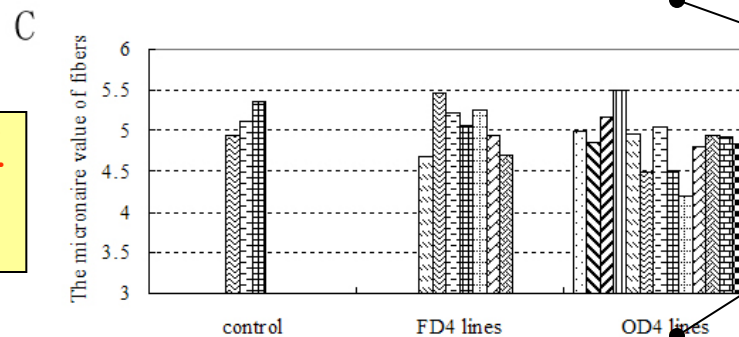
Fiber length of OD4-41 increased by 17.14%



13.2%

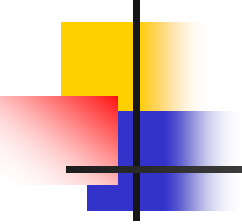


A) Fiber length (mm);
B) Fiber strength (cN/tex);
C) micronaire.

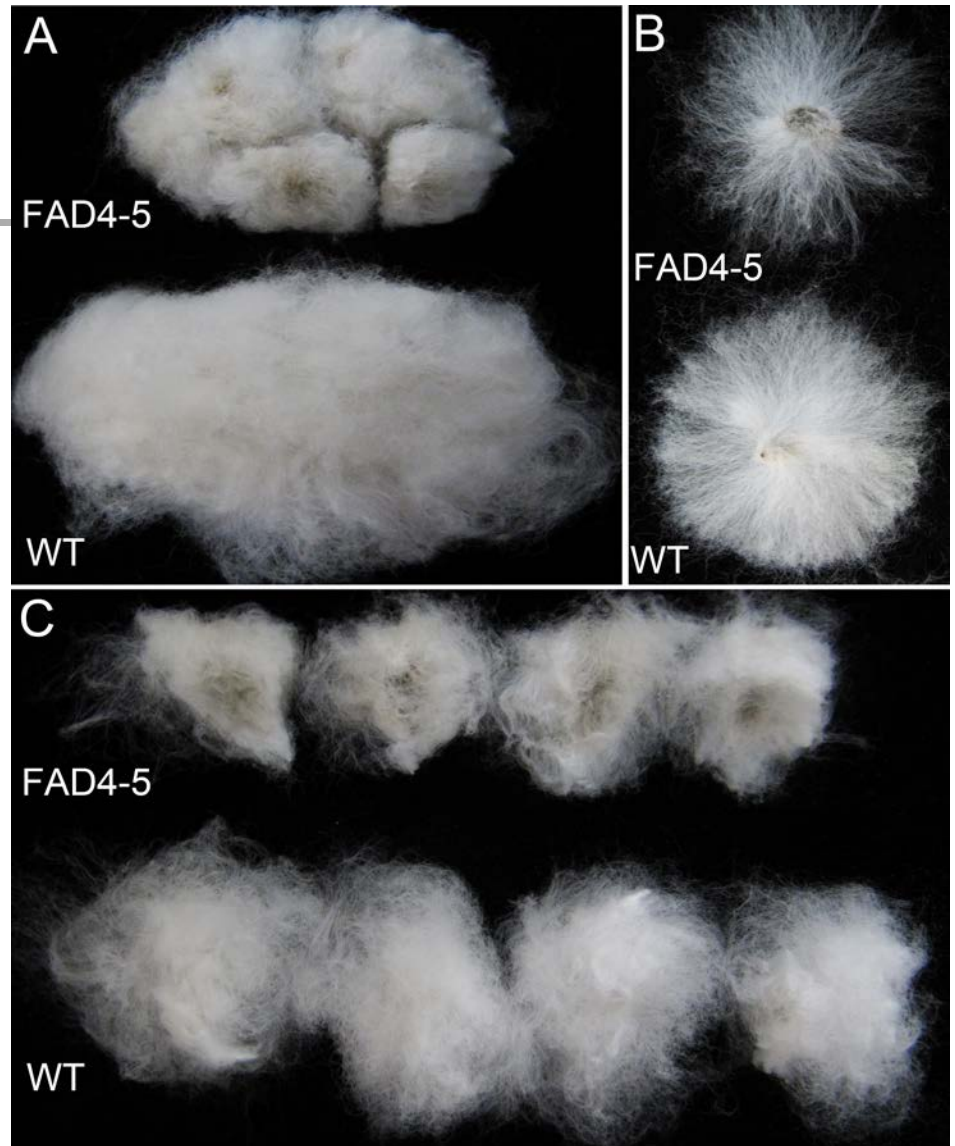


16.7%

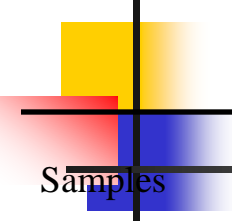
14.4%



**Inhibit the GhDWF4
expression result
in the decline of
fiber number and
length**

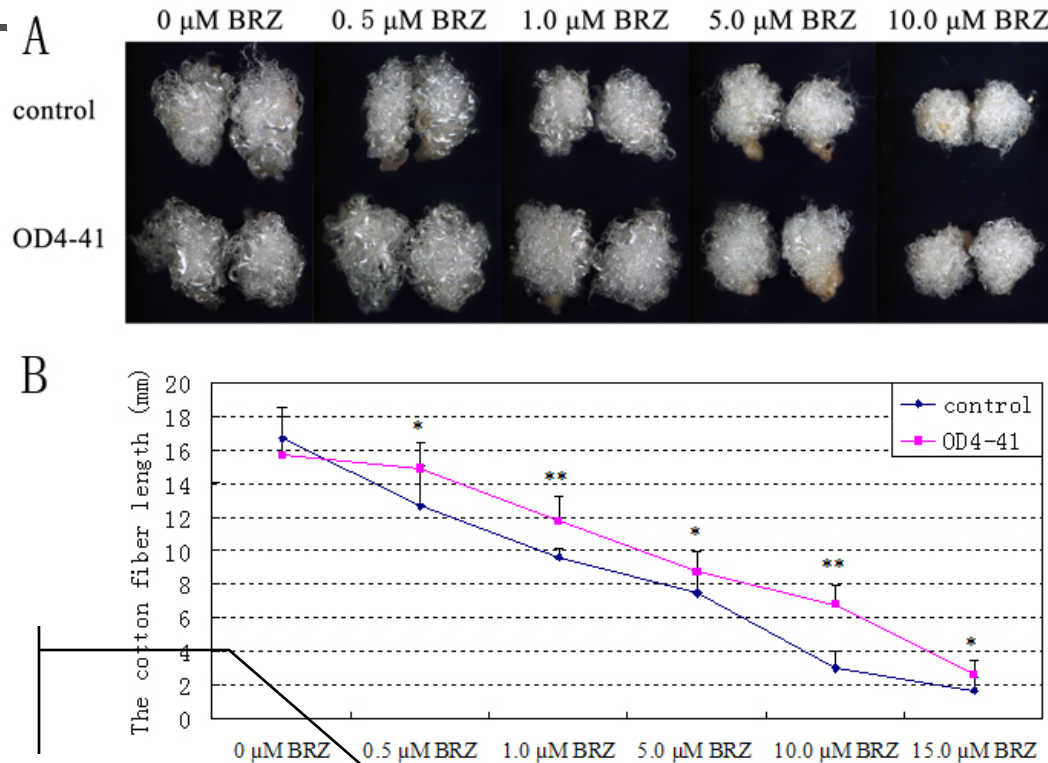


Fiber quality report in 2010



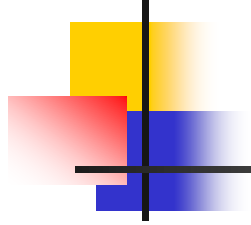
| Samples | Upper half mean length (mm) | Uniformity (%) | Micronaire | Elongation (%) | Strength (cN/tex) |
|---------|--------------------------------|------------------|-----------------|-----------------|-------------------|
| control | 28.24 ± 0.45 | 82.76 ± 0.97 | 6.28 ± 0.09 | 6.78 ± 0.10 | 27.41 ± 0.75 |
| OD4-8 | 32.1 | 84.3 | 4.8 | 7.1 | 31.1 |
| OD4-11 | 30.53 ± 0.28 | 85.33 ± 0.53 | 5.15 ± 0.19 | 6.83 ± 0.05 | 30.78 ± 1.00 |
| OD4-18 | 31 | 85.4 | 4.5 | 6.7 | 28.2 |
| OD4-26 | 31.55 ± 2.76 | 84.6 ± 1.56 | 5.5 ± 0.99 | 6.95 ± 0.07 | 29.25 ± 0.78 |
| OD4-30 | 32.2 | 85.7 | 4.5 | 6.9 | 30.5 |
| OD4-83 | 30.85 ± 1.37 | 85.33 ± 0.88 | 5.12 ± 0.65 | 6.83 ± 0.08 | 30.2 ± 1.37 |
| OD4-166 | 30.75 ± 1.91 | 84.02 ± 0.50 | 4.78 ± 0.66 | 6.77 ± 0.10 | 29.28 ± 0.70 |

Fiber elongation was repressed when GhDWF4 activity was inhibited



Specific inhibitor
of DWF4 protein

The effects of applied **brassinazole (BRZ)** on cotton fiber elongation.
A) The growth condition of ovules and fibers treated by gradient BRZ.
B) The length of fibers treated by gradient BRZ.



Other genes involved in the down-stream pathway were functional characterized in fiber cell growth and development

GhROT3 is a homologue of C23- and C3-hydroxylation enzyme

A

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GhROT3 : M A L Y I A K R E G F E S K E D M ----- G L I I I S L M G V V F W Y --- K M K M M R R E D I G I N - K S G S G S G G G V P R C N D G W P I G E T I F I A S G Y S S C P V S F M K R R L I H G S - V F K T I L I G T F : 107
AtROT3 : M P P A S A - G L F R S P E N L F W P Y N Y M D Y L V A G F L V I T A G I L L R F L W L --- R L R N S K T D G D E E E D N E E K - K G G V I P N G S G W P I G E T I N F I A C G Y S S P V I F M K R K S I Y G S - V F K T I I G T F : 117
OsROT3 : M V S A A A G W A A ----- P A F A V A A V V I ----- V V L C S E L L R R R R G A G S G K - - G D A A A A A R L P G S G W P I G E T I F V S G A Y S P R P E F V I K R R K I H G S A V F R S I I G S A : 98
PsROT3 : M --- D T T W I L ----- F A T P ----- I F L C T L L Y R N L S L K L K - - S K H G N - - C L H G S L G W P I G E T I F V S G A Y T D R P E F F M K R R I Y G S - V F K S E I I G S T : 85
PtROT3 : M ----- E Q K ----- - - - - - S G V L P G S G W P I G E T I F I A A G Y S C P V S F M K R R S I Y G S - V F K T I I I G T F : 56
RcROT3 : M --- C W ----- V V M G A I V I G W C F Y S N N K M K M K M N S K V K G M K S M G K S S V P P C N I G W P I G E T I F I A C G Y S C P V N F M K R K S I Y G S - V F K T I I I G T F : 95
VvROT3 : M S V I H W C W ----- V V V G V V M G - W C W Y - - K S I K N K A E E - - - - - G S V P P C N I G W P I G E T I F I A S G S S F P V S F M K R K S I Y G S - V F K T I I I G T F : 82
    
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Anchor region

Profine

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GhROT3 : I I V S I D P V N K V V L C N H G N T F E A Y P K S I T E I L G E S S I I O M N G L K R L H A L I A G F I R S P Q R K A R I T R D I E R Y V K L I L A S R K D Q I V V Q D V R K I T F F V L V R V L V S I T P C E L I N F M K R E E E I : 232
AtROT3 : I I I S I D A E V N K V V L C N H G N T F E A Y P K S I T E I L G E S S I I S I N C P H O K R L H T L I G A F I R S P H U K O R I T R D I E A S V I T I A S A Q L P I V V Q D I K K M I F E I L V K V L M S T S P C E M N I L K E F F E I : 242
OsROT3 : I V V T I D A E ----- P R R V H L E G A F K S S H U K S C L T A M R R R L S P A L S E F D G S L I V C H L A K S V F E I L V R S L I G E A C E P M O Q L K O F F E I : 185
PsROT3 : I I V S I D A L V N K E I L S D A K V E F E S Y P K S I M K M G E S S I I L I N G I L R R R I H L I G A F I R S Q U K I L I T T E M K Y V Q E S M A N M E D Q I V I Q D I K K I A F F V L V K L I S I D P C E M E L I K E F F E I : 210
PtROT3 : I I V S I D P A V N K V V L C N H G N T F E A Y E K S V R E I E E Y S I I Q T N G I L K K V H L I G F I R S P O B K T I T R D I E H H V K L I L A S R K D Q I V V Q D I F L V C E T C F E F V L V K L I S I D S D L D L K R E F F E I : 181
RcROT3 : I I V S I D P V N K V V L C N H G N V F E I A Y E K S I R E L L G E S S I I O V N G N L K K L H V I A G F I R S P Q R K A I T R D I E N S V K L I L A S R K G M F V V Q D I K K I T F F V L V K L M S T S P C E N I L D L K R E F F E I : 220
VvROT3 : I I V S I D P E V N K V V L C N I G N V E I F A Y H Q I T E I L G E S S I I O M N G S L K R V H A L I G G F I R S P Q R K A R I T R D I E R Y V K L I L A S R K D Q I V V Q D V R K I T F F V L V R V L V S I T P C E L I N F M K R E E E I : 207
    
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GhROT3 : E G L I L P I K I P G T R L Y S L A K E R L K V V K I V E R K L A N E --- E E T D E K S M A K D A V D V L L D S S E E P S D Q I K Q S L P L S I S G N T I E M M I P E E T V P V M T L A K F L S D Q P A L C L A E E N M I : 354
AtROT3 : E G L I L P I K I P G T R L Y S L A K E R L K M V K I V E R R V A M T T T --- S P A N D V D V L L R E G D S E - - - - - K Q S O P S E V S G K I V M M I P E E T M T A M T L A K F L S D N P A L A K E E N M M : 355
OsROT3 : V G I M S L P I K I P G T R L Y S L A K K M A R L I R I E K A R R A A A S - - - - - P E R D A I D V L I N G S D E - - - - - L T D E I I A D N M I L M I P E E S V E I I T L A K F L S E P A L A K E E N I : 294
PsROT3 : E G L M S L P I S I P G T K L Y S L A K K M A K L V K I I P A R N K - G V T E - - - - - V P R D V D V L L N T S E K - - - - - L T I L I A D N I I E M M I P E E S V E I I T L A K Y L S E P A L A K E E N I : 318
PtROT3 : E G L I L P I K I P G T R L Y S L A K R I E K L V K I V E R K L G E K S E D Q E H O N T I P D A V D V L L E I S G E Q N D - - - - - K Q S L S D Q I I S H I E L M I P E E T V P M S M T L A K F L S D Q P A L E L L E E N M I : 303
RcROT3 : E G L I L P I K I P G T R L Y S L A K E K L K M V K I V E R K F A M E K C N D E A N L R - - - - - P N E I L D V L L R E N V E G N D H K Q S - - - - - D I S S N I V M M V P E E T V P M A M T L A K F L S D Q P A L K M E E N M I : 337
VvROT3 : E G L I L P I K I P G T R L Y S L A K E R L K L V R I V E R R A A M D R G D H E T - L K G P P N D A V D V L L R E T G D S S E T S R - - - - - L S L I S S N I E M M I P E E T L E T A M T L A K F L S D Q P A L N C L A E E N M I : 327
    
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Domain A

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GhROT3 : K K K R E G G E D Y S W T D Y S I P E T C N V I S E T L A M N I N G W R K A L K D V E I K G L I P G W V V T S F S V H M D E E N Y E N P F P P W R W E T I - G A A A N N S F I P F G G G Q R L C P G L S L R L E S I F L H H I : 478
AtROT3 : K R K R E L G E E Y S W T D Y S I S E T C N V I N E T L A M N I N G W R K A L K D V E I K G L I P G W V V A S F S V H M D E D I Y D N P F P P W R W E T I N G S A N S S I G E T P F G G G Q R L C P G L S L R L E S I F L H H I : 480
OsROT3 : K R K R T D M G E T L O W T D Y S I S E T C H V I T E T L A N I G G I M R K A V R D V E K G L I P G W V F V F S V H L D T T I D P P F P P W R W - E K D M S - - - - - N G S E T P F G G G Q R L C P G L S L R L E S I F L H H I : 416
PsROT3 : K K K Q Q L E E F L O W S D Y S I P E T K I I T E T L A M N I N G W R K A L K D V E I K G L I P G W V V A F S S V H L D E K N Y D G P F P P W R W - O E K D M N L S N F S P F G G G Q R L C P G L S L R L E S I Y L H H I : 442
PtROT3 : K R K R D S G E D Y S W T D Y S I S E T C N V I N E T L A M N I N A W R K A L K D V E I K G L I P G W V V T S F S V H M D E E N Y E N P F P P W R W E T - G A S V K N N F P F G G G Q R L C P G L S L R L E S I F L H H I : 427
RcROT3 : K R K R D T C D E Y S W T D Y K - - - - - I V V D I Y S K - - - - - S N D F S C L I P G W V V A S F S V H M D K E Y E N P F P P W R W E T - G T A V N N S C F P F G G G Q R L C P G L S L R L E S I F L H H I : 443
VvROT3 : K K K A L S G E D Y S W T D Y K I S E T C N V I N E T L A M N I N A W R K A V K D V E I K G L I P G W V V A S F S I H M D E E N Y E N P F P P W R W E A - A S V N N N S Y P F G G G Q R L C P G L S L R L E S I F L H H I : 451
    
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Domain B

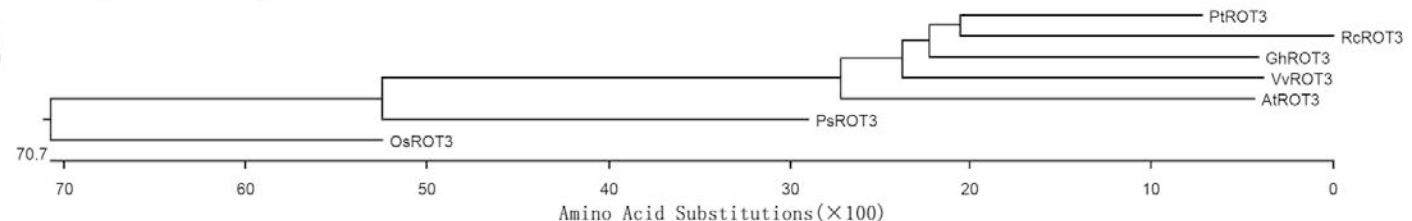
Domain C

Heme binding

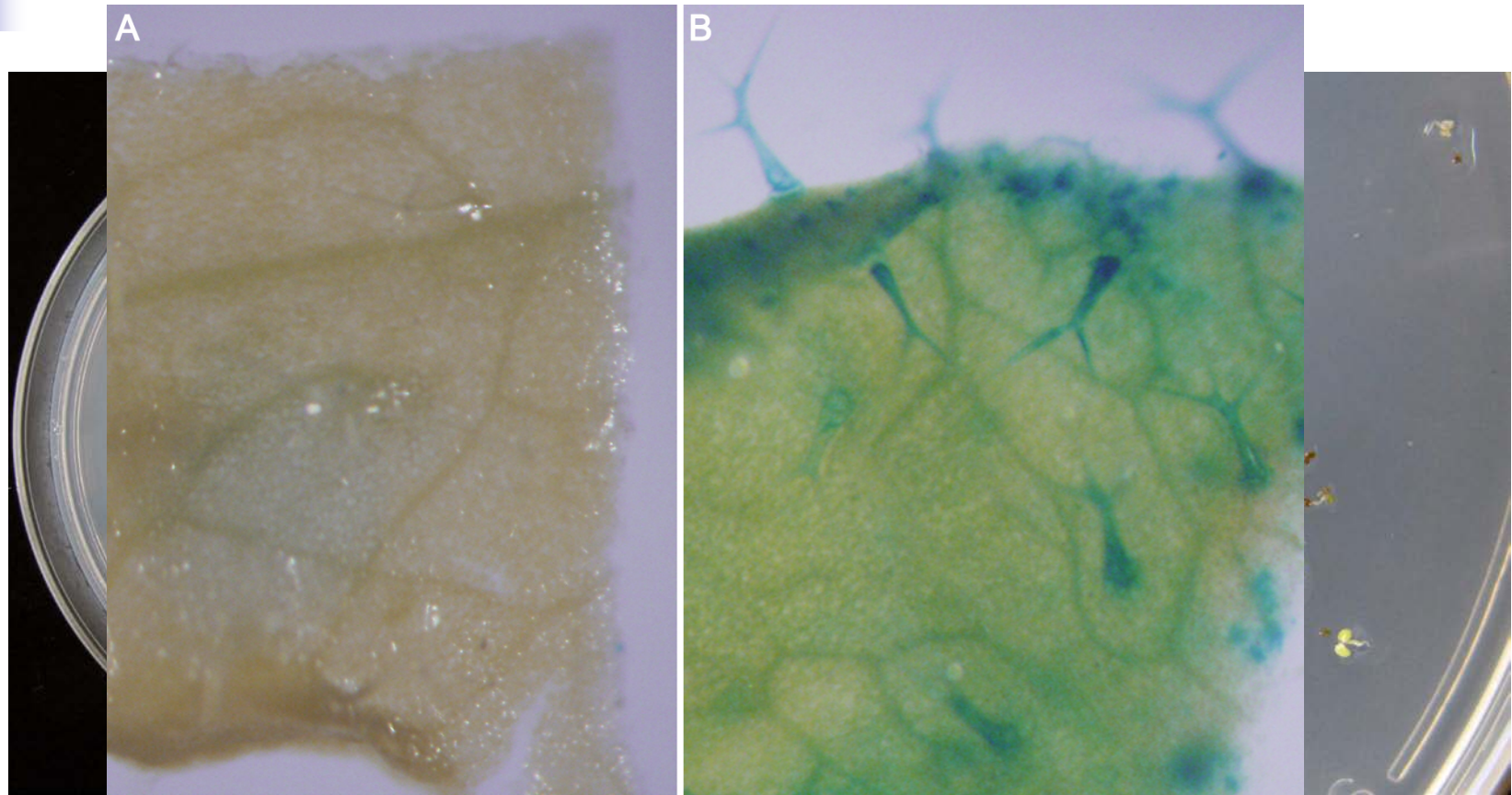
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GhROT3 : V T I Y R V A K D I I V F P T V K M K K L P I V T P L C - N Y - - - - - Q : 514
AtROT3 : V T I Y S I A S E D I V F P T V K M K R L P I V A T V D D S A S P I S L E D H : 524
OsROT3 : V T S F R V A S E D I V N F P T V R L K R E M P I V I A K E D - - - - - D D : 452
PsROT3 : V T S F R V A S E D I V N F P T V R M K R E M P V I V - - K R V - - - - - E I : 476
PtROT3 : V T D Y R V A K D I I V F P T V K L K K L P I V T S I G - N S - - - - - S : 463
RcROT3 : V T D Y R V A K D I V F P T V K M R R L P I V T I L H Q P Y - - - - - Q : 480
VvROT3 : V T D Y R V A K D I V V F P T V K M R K L P I V T P L - - - - - P P - - - - - L : 486
    
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B



GhROT3 rescued the *rot3* mutant

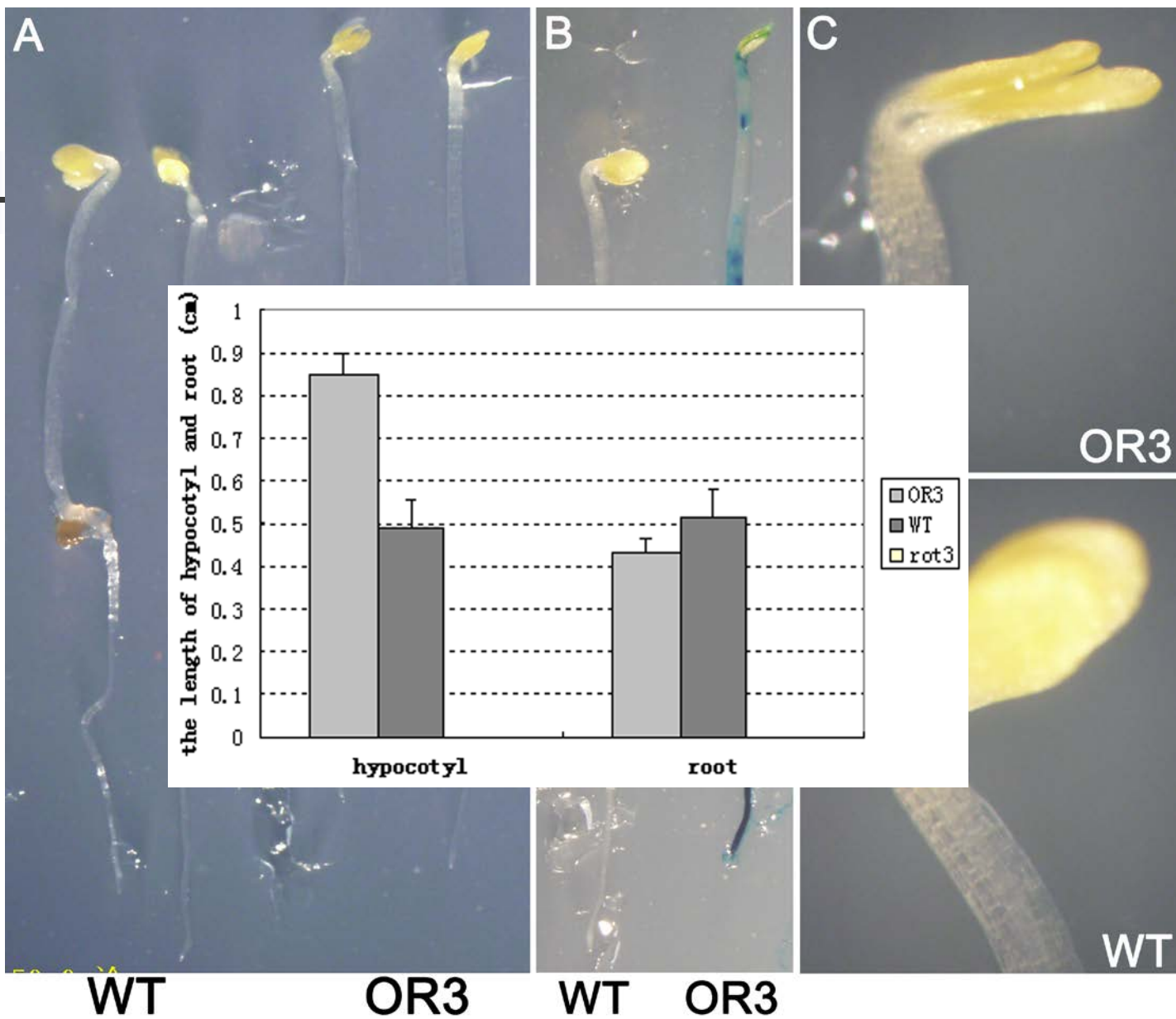


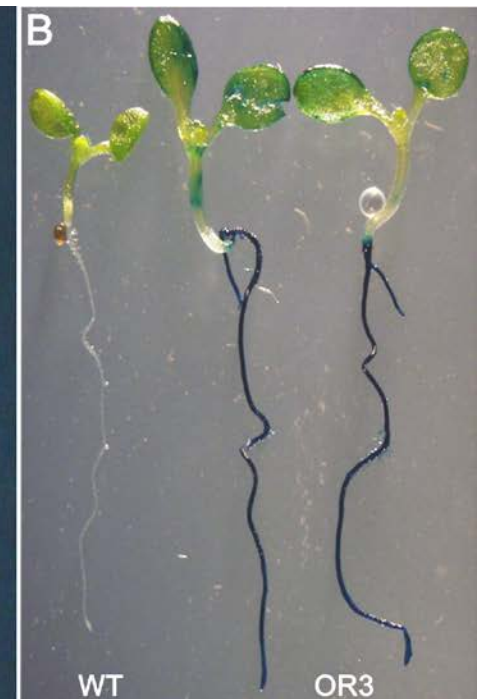
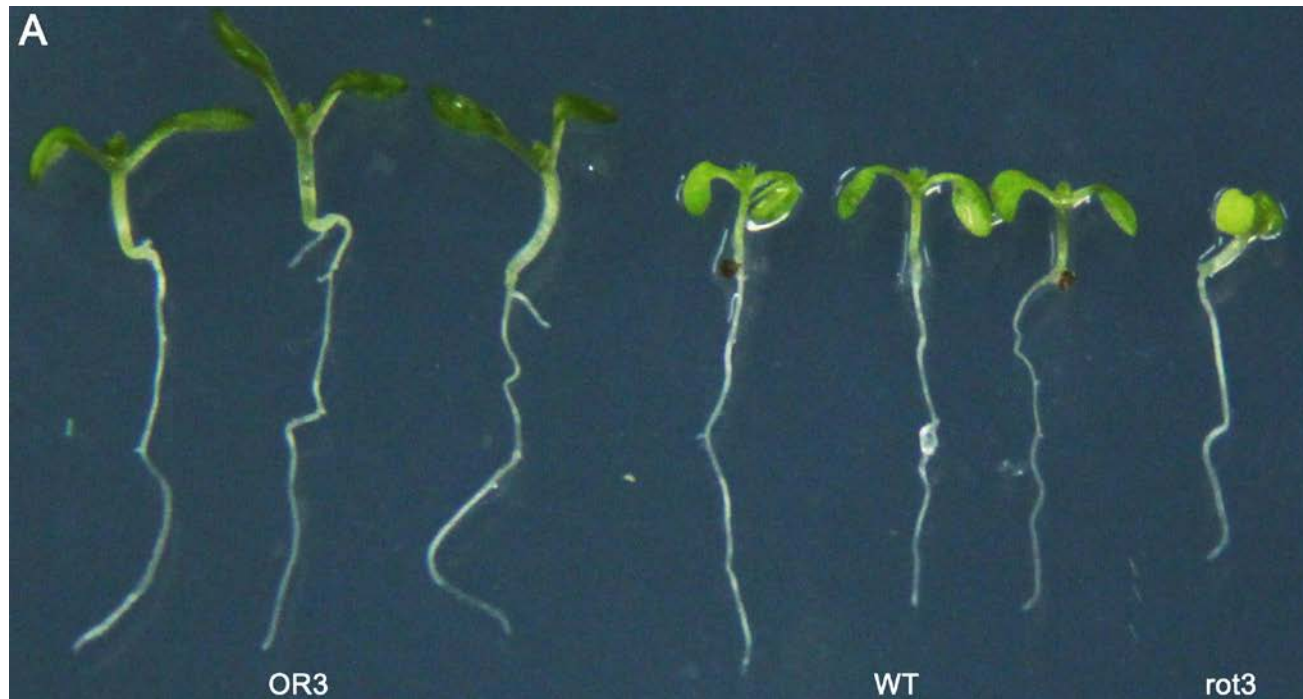
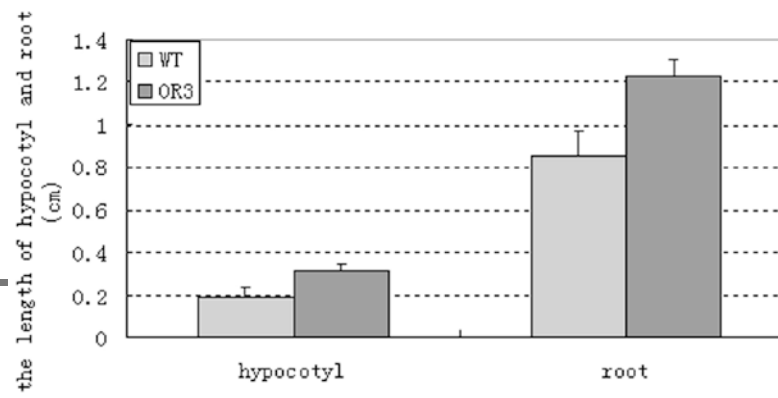
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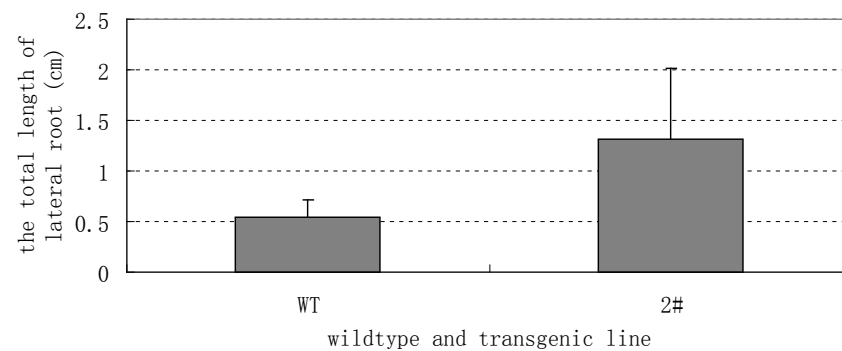
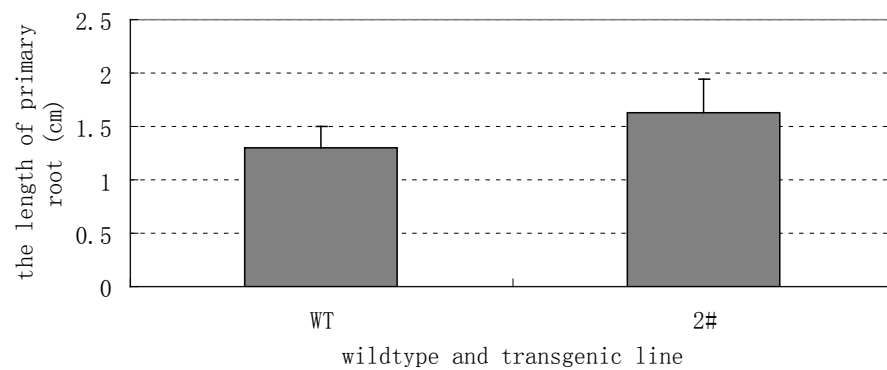
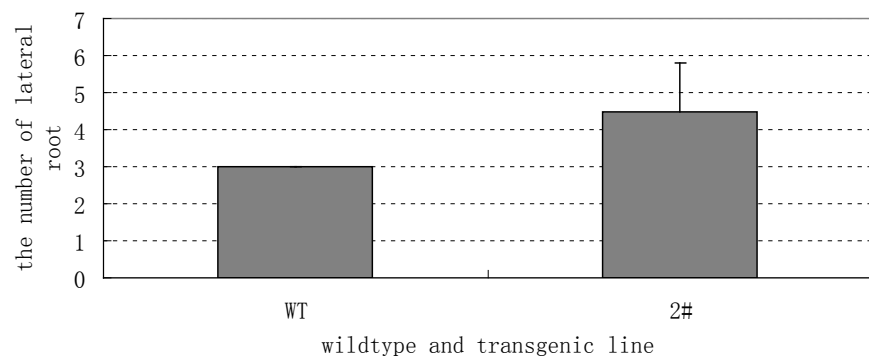


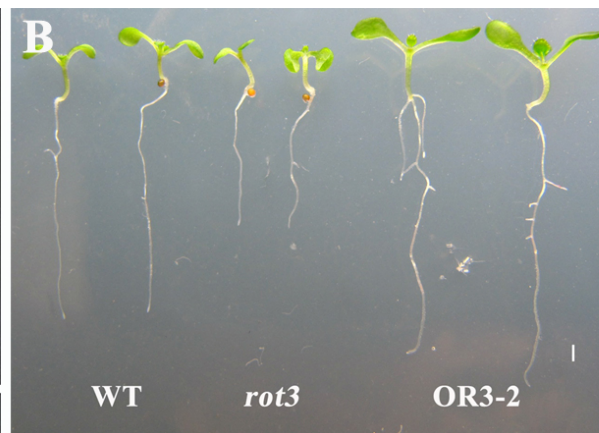
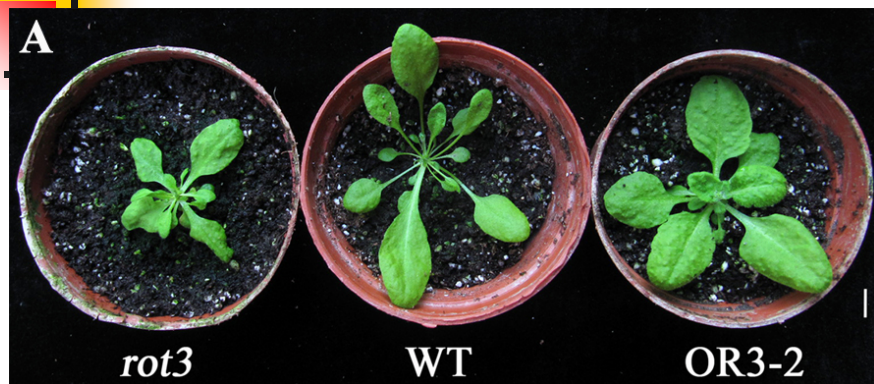
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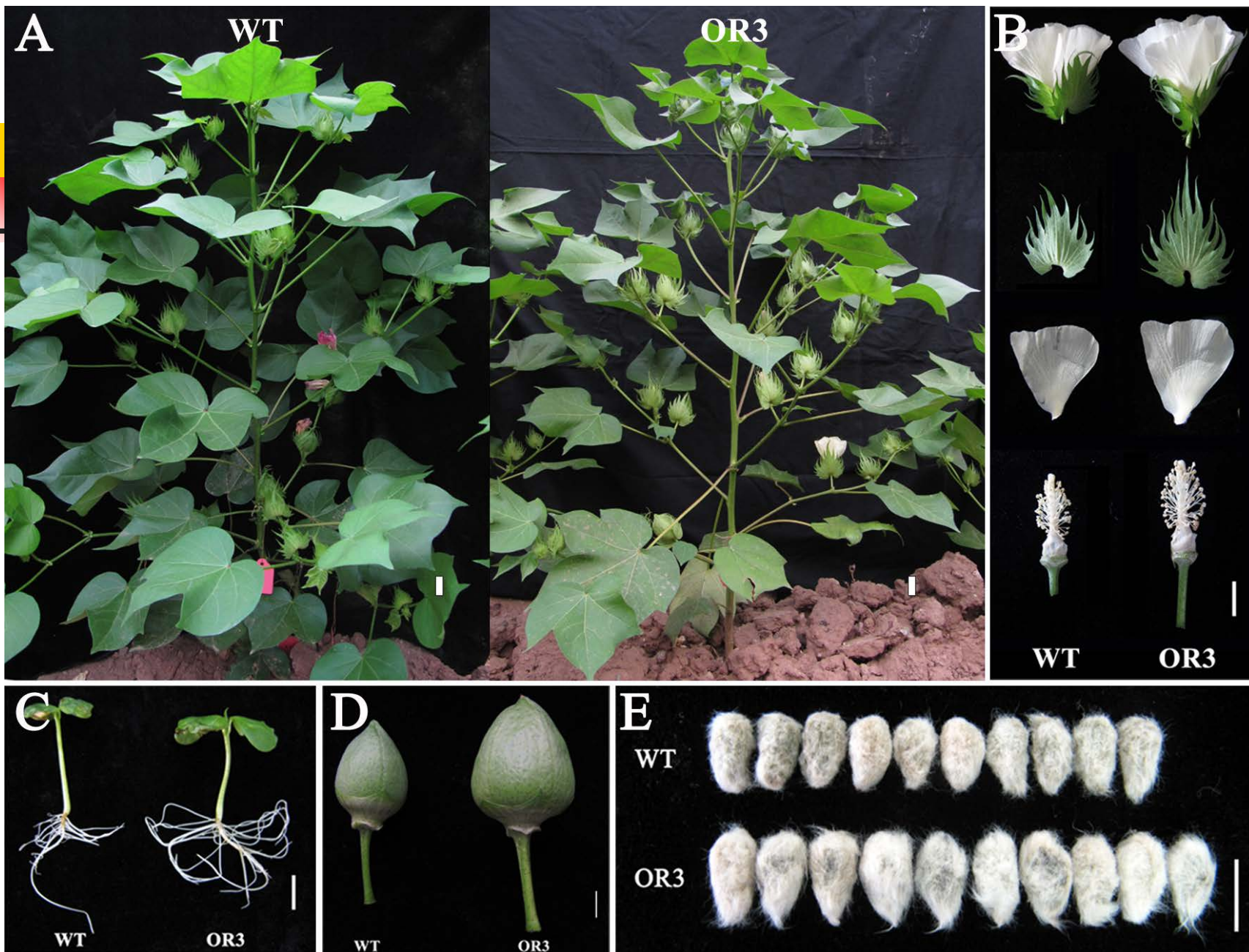


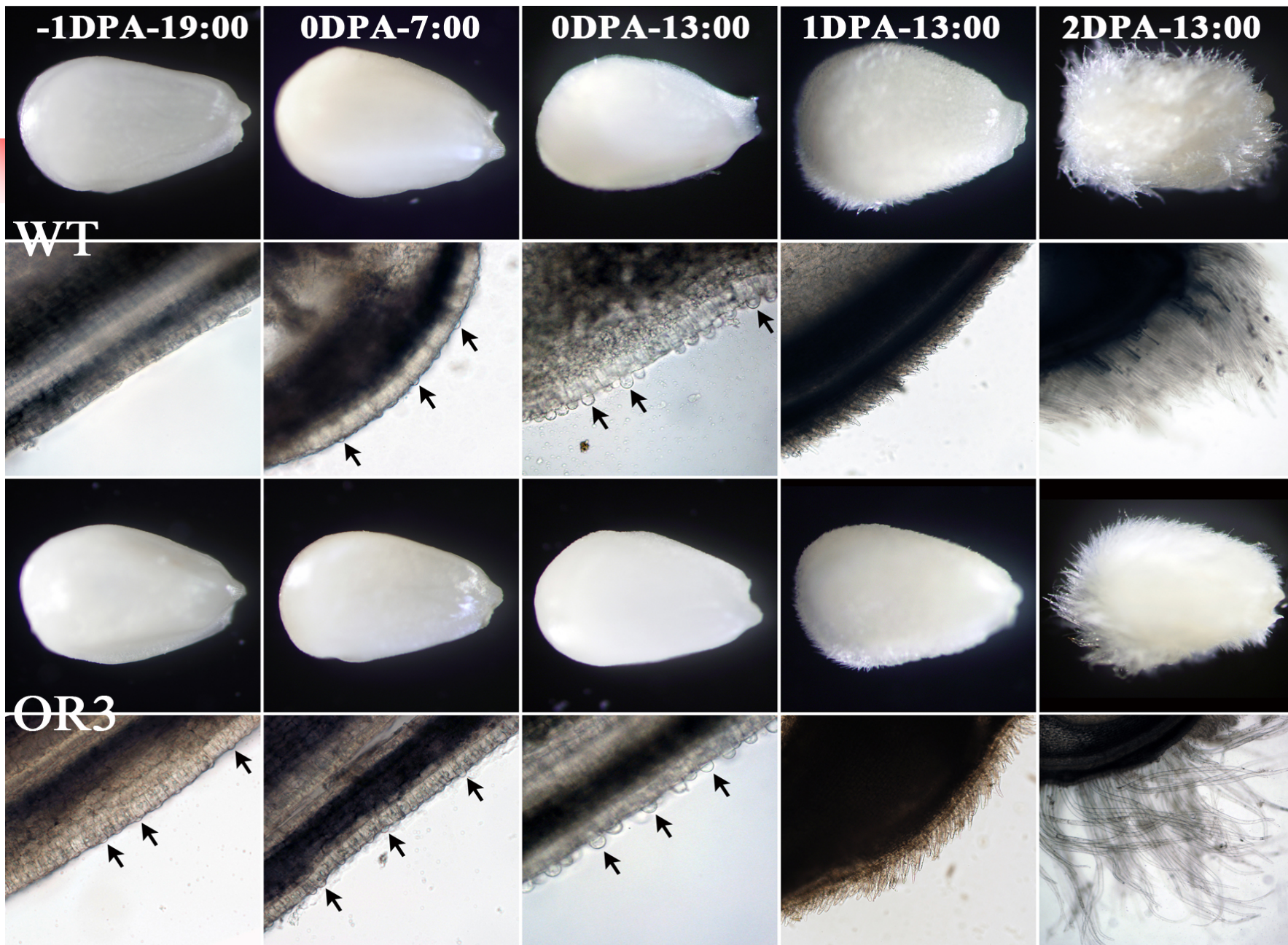


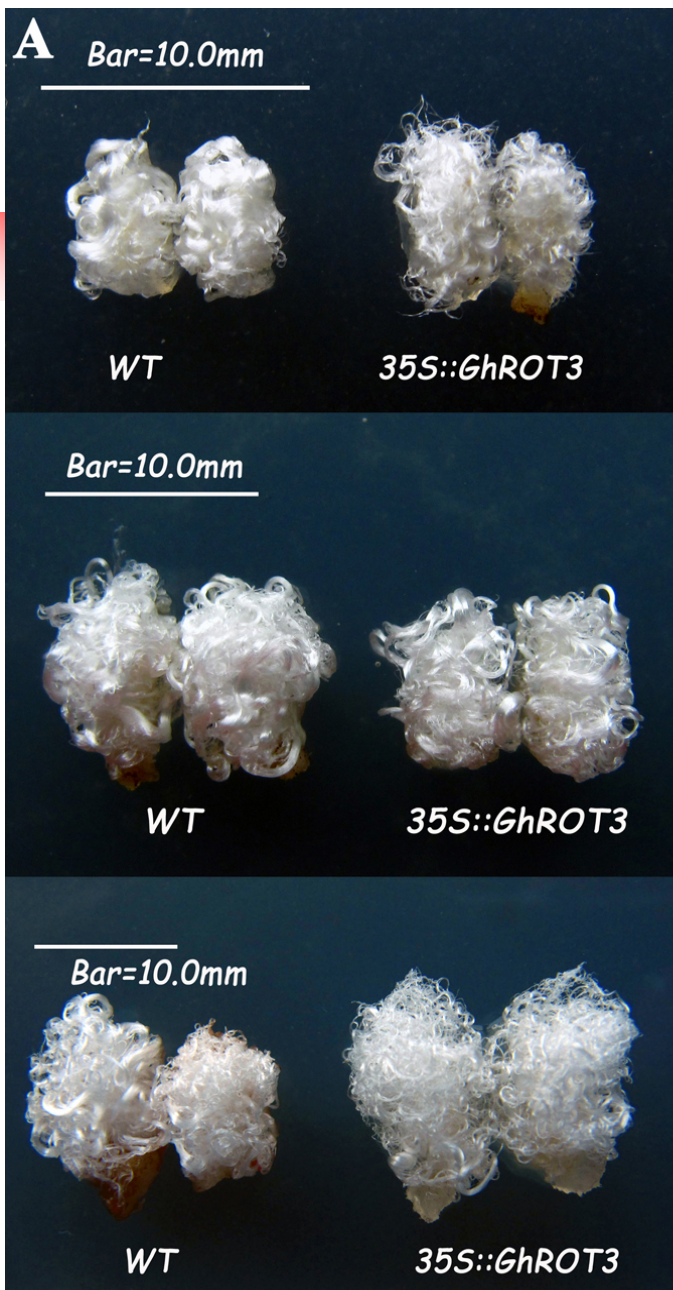








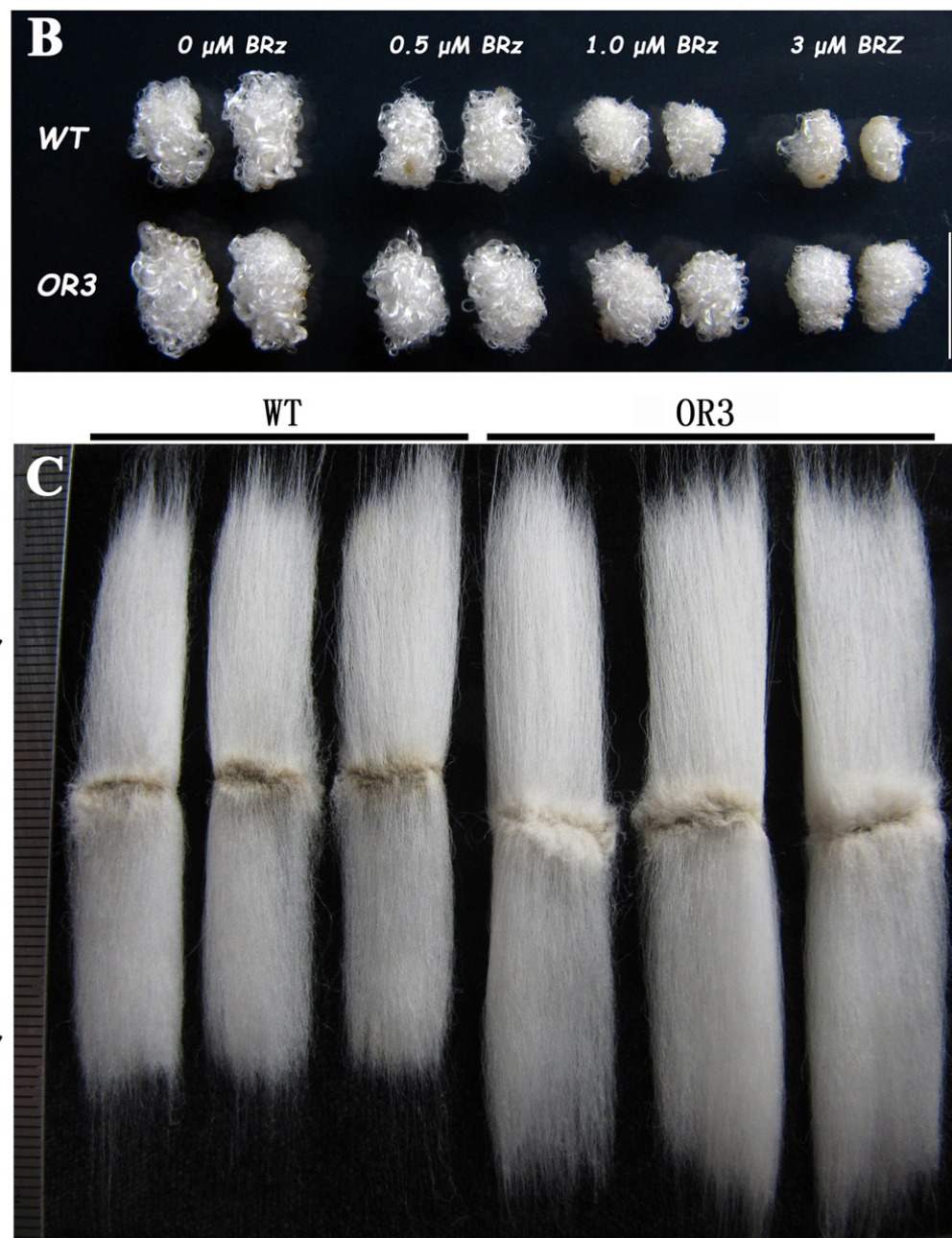




5-day

10-day

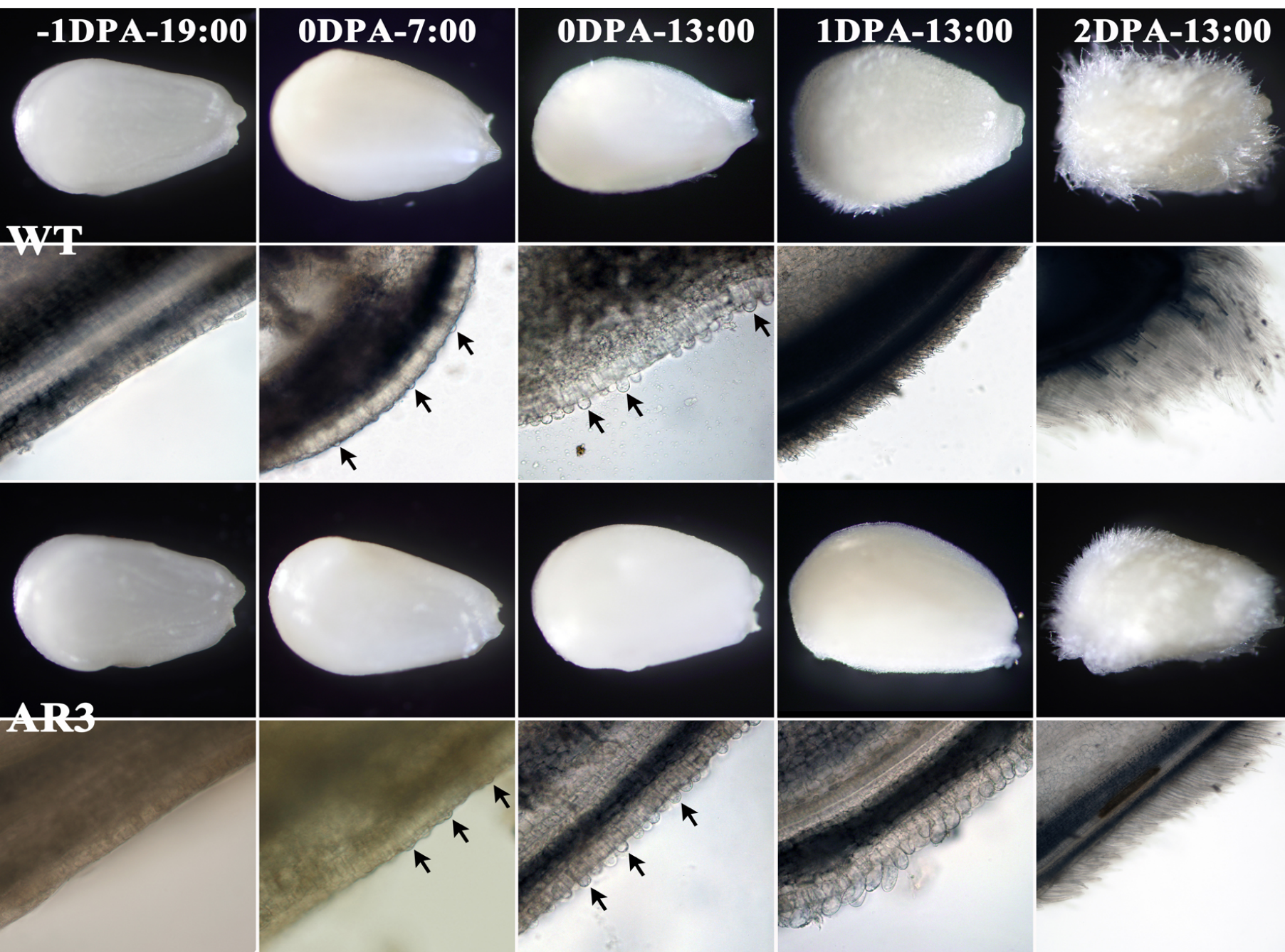
15-day



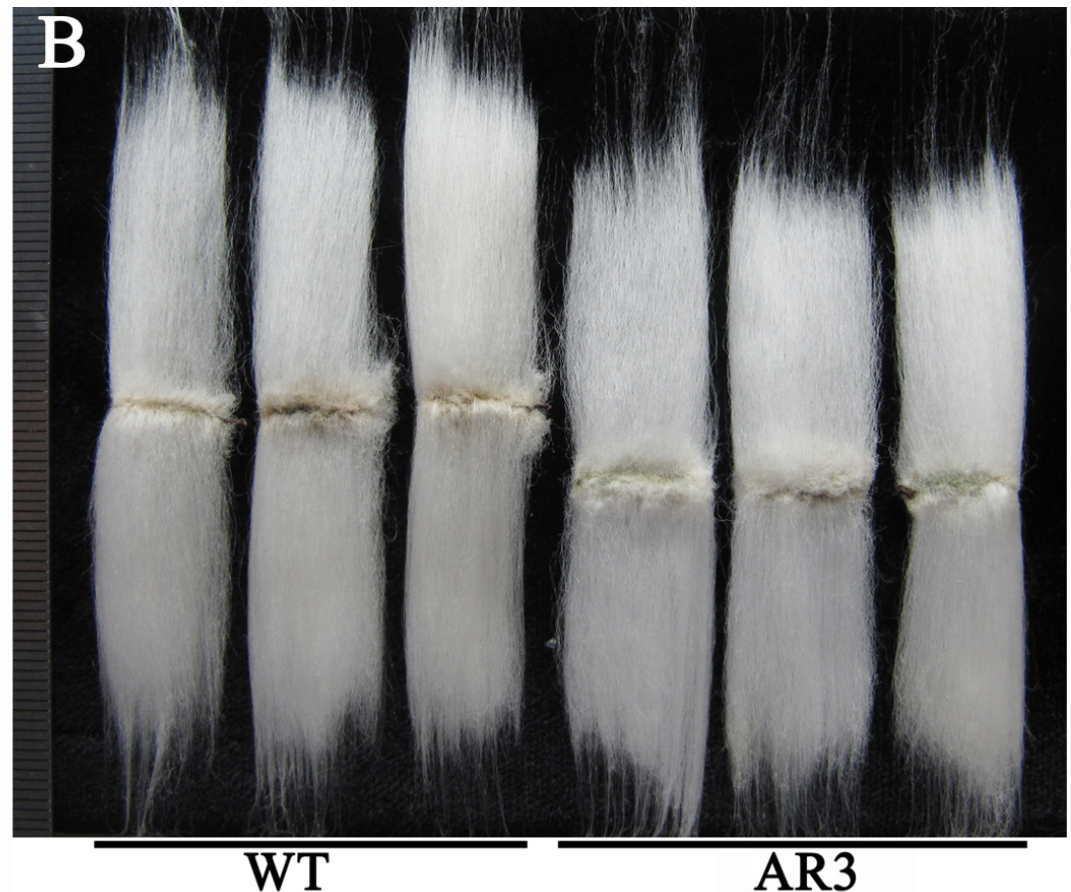
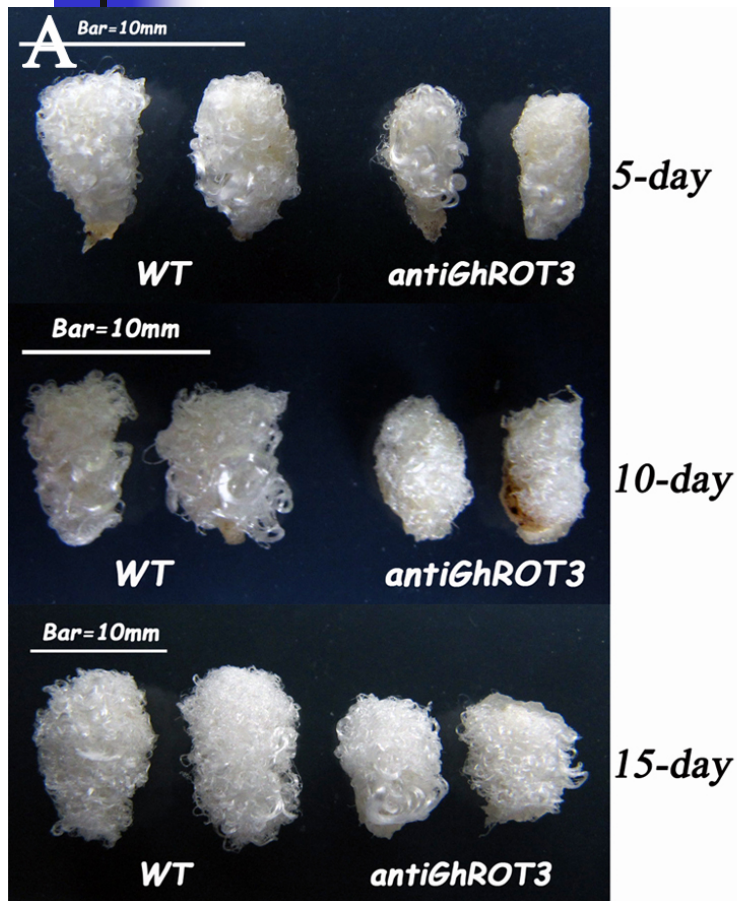


■ Overexpressing GhROT3 plant

suppressing GhROT3 plant



Decreased *GhROT3* expression suppressed fiber cell elongation

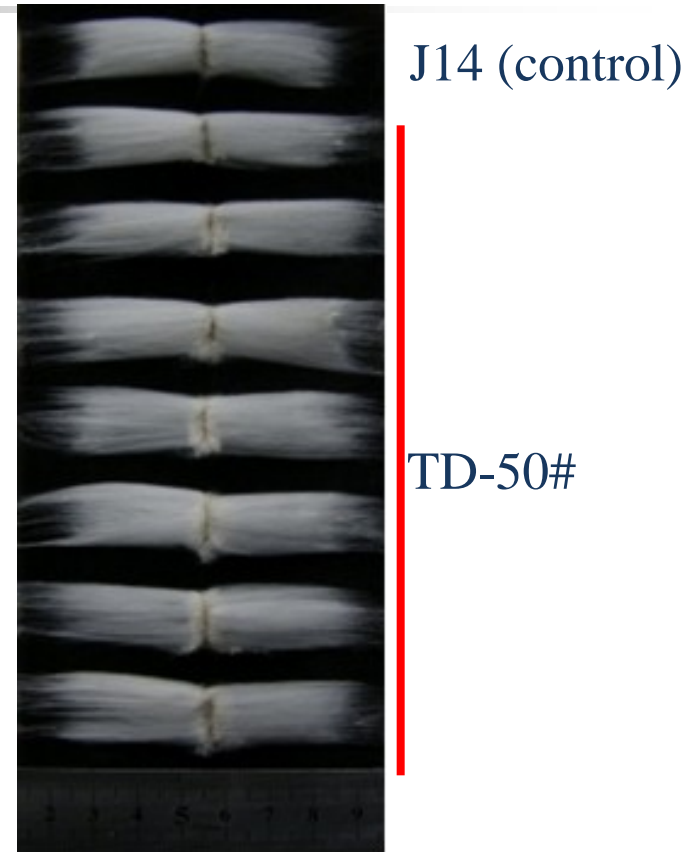
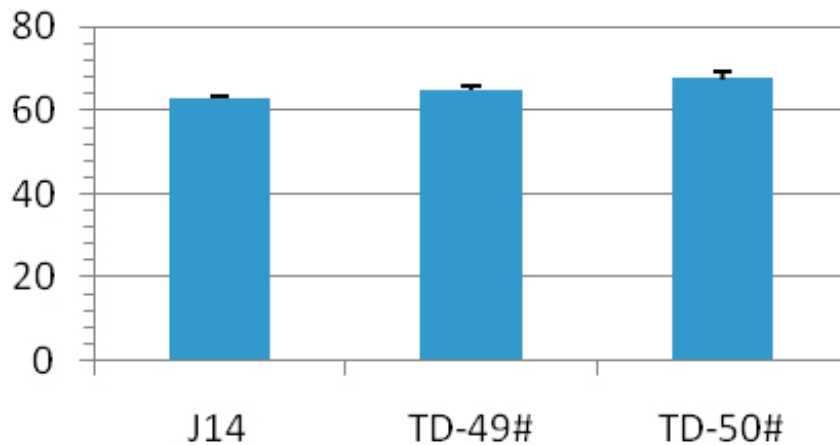




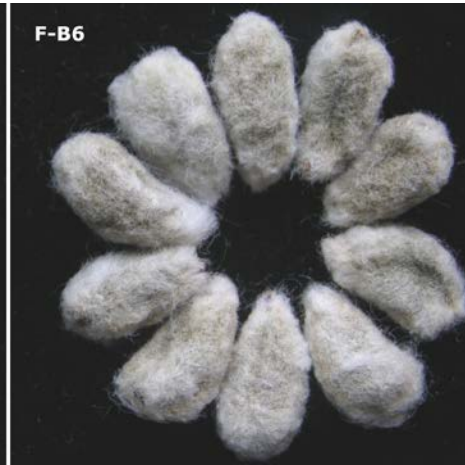
Fiber quality report in 2011

| samples | Upper half mean length (mm) | Uniformity (%) | micronaire | Elongation (%) | Strength(cN/tex) |
|---------|--------------------------------|----------------|------------|----------------|------------------|
| OROT3 | 33.03 | 86.90 | 4.72 | 6.10 | 32.5 |
| CK1 | 30.15 | 85.90 | 4.66 | 6.40 | 29.8 |
| CK2 | 28.49 | 85.30 | 4.43 | 6.60 | 28 |

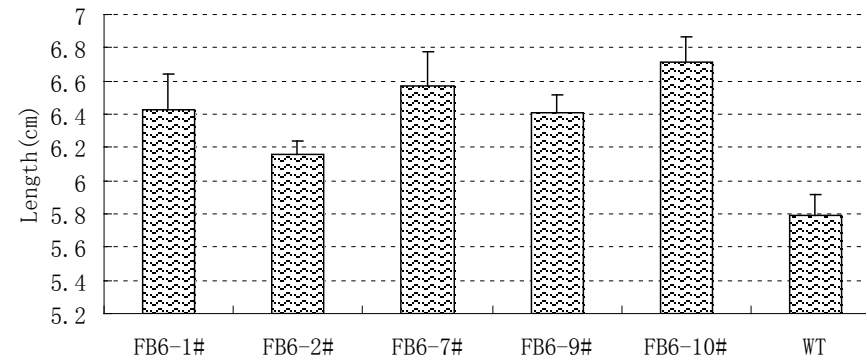
Overexpressing GhCPD promoted fiber cell elongation



PFBP7::GhBR6OX2 increased the seed size and fiber length of transgenic cotton



Fiber Length of Transgenic Cotton and Wild-type Cotton



Overexpression of *GhDWF4* increases the bioactive BRs in cotton fibers

| BRs (ng/g FW) | control | | OD4-41 | OD4-16 |
|------------------|--------------|--------------|--------------|--------------|
| | 12dpa fibers | 20dpa fibers | 12dpa fibers | 20dpa fibers |

Late C-6 oxidation

The product of DWF4 protein

| | | | | |
|------------|------|------|------|------|
| 6-DeoxoCT | 0.44 | 0.61 | 23.7 | 28.1 |
| 6-DeoxoTE | 0.01 | 0.01 | 0.01 | 0.22 |
| 6-Deoxo3DT | 0.04 | 0.06 | 0.05 | 0.07 |
| 6-DeoxoTY | 2.04 | 1.92 | 5.22 | 12.9 |
| 6-DeoxoCS | 0.08 | 0.18 | 0.29 | 4.42 |

← About
50 folds

← 2.5-24.5
folds

Early C-6 oxidation

| | | | | |
|-----|------|------|------|------|
| CT | nd | nd | nd | nd |
| TE | 0.08 | 0.07 | 0.07 | 0.06 |
| 3DT | nd | nd | nd | nd |
| TY | 0.31 | 0.11 | 0.35 | 0.12 |
| CS | 0.06 | 0.05 | 0.07 | 0.05 |
| BL | nd | nd | nd | nd |

The content of bioactive BRs in developmental cotton fibers (nd: not detected).

Other phytohormones were regulated in *GhDWF4*-overexpressing fibers

| Phyto-hormones | Change of content | Regulated gene analyzed by Q-PCR | DGE analysis by solexa technology |
|----------------|---|---|--|
| IAA | Reduced by 35.4% | <i>GhABP1</i> was down-regulated | <i>SAUR</i> genes were down-regulated |
| GAs | GA1 reduced by 18%, GA4 increased by 11% | <i>GhGA20OX3</i> was down-regulated | <i>DELLA</i> was up-regulated <i>GID</i> was down-regulated |
| ABA | Reduced by 33.8% | <i>GhABA1</i> , <i>GhNCED2</i> were down-regulated | |
| ethylene | | <i>GhACO</i> , <i>GhACS</i> were down-regulated | <i>GhACO1</i> and <i>GhACO2</i> were down-regulated |



Digital gene expression (DGE) by solexa sequencing

Tutor related genes were up-regulated

Aquaporin: Plasma membrane Intrinsic Proteins (PIPs) and Tonoplast

Intrinsic Proteins (TIPs)

Sucrose transporters and sugar transporters

Cell wall biosynthesis related genes were up-regulated

Cellulose synthase and keratin

Other genes related to fiber growth were regulated

Up-regulated

CaM and CDPK related kinase

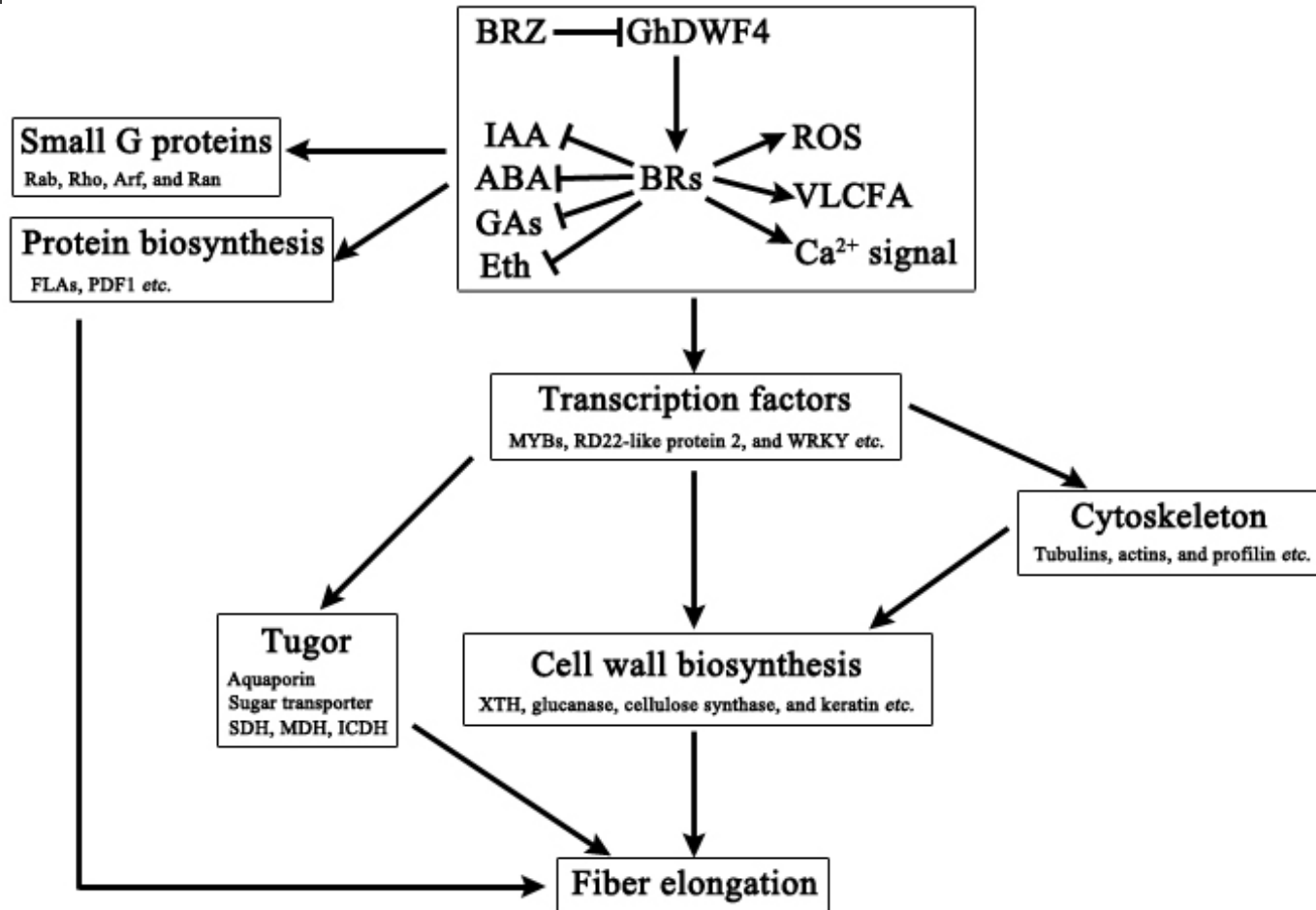
Fasciclin-like arabinogalactan proteins (FLAs) and PDF1

Down-regulated

Peroxidase and catalase

Cytoskeleton related genes

The hypothetic model of *GhDWF4* promoting fiber elongation





Acknowledgement

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Thank you!