Comprehensive speed breeding: a high-throughput and rapid generation system for long-day crops

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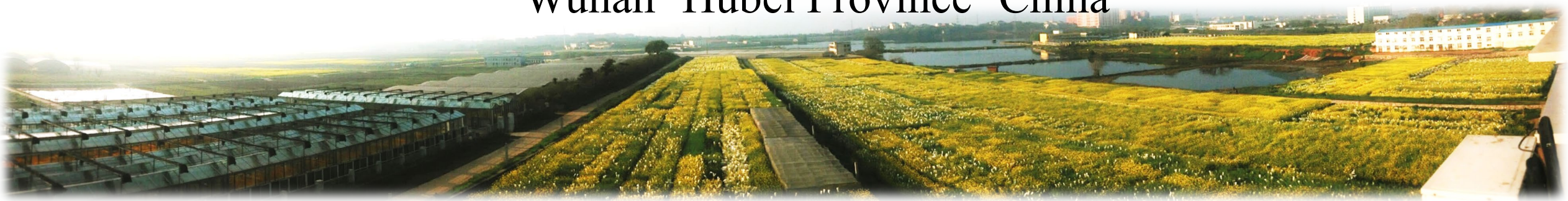


outline

- Accelerated generation advancement is increasingly important for crop improvement
- Comprehensive Speeding Breeding (CSB) : a high throughput system for rapeseed
- CSB is powerful for QTL pyramiding
- CSB shows great potential for rapeseed functional genomics research

Part I

Accelerated generation advancement is increasingly important for crop improvement

Higher genetic gain is expected for modern breeding

https://www.theworldcounts.com/populations/world/people

Table 5.1 Increase in agricultural production required to match projected demand, 2005–2050 (percent)

	2005/07	2050	2005/07 2012	2013-2050
World				
As projected in AT2050	100	159.6	14.8	44.8
With updated population projections (UN, 2015)	100	163.4	14.8	48.6
Sub-Saharan Africa and South Asia				
As projected in AT2050	100	224.9	20.0	104.9
With updated population projections (UN, 2015)	100	232.4	20.0	112.4
Rest of the world				
As projected in AT2050	100	144.9	13.8	31.2
With updated population projections (UN, 2015)	100	147.9	13.8	34.2

Source: FAO Global Perspectives Studies, based on UN, 2015, and Alexandratos and Bruinsma, 2012.

FAO 2022

Human population VS Genetic gain

How to balance the human population and food supply?

Breeder's Equation

Selection intensity

Selection accuracy

Genetic variance

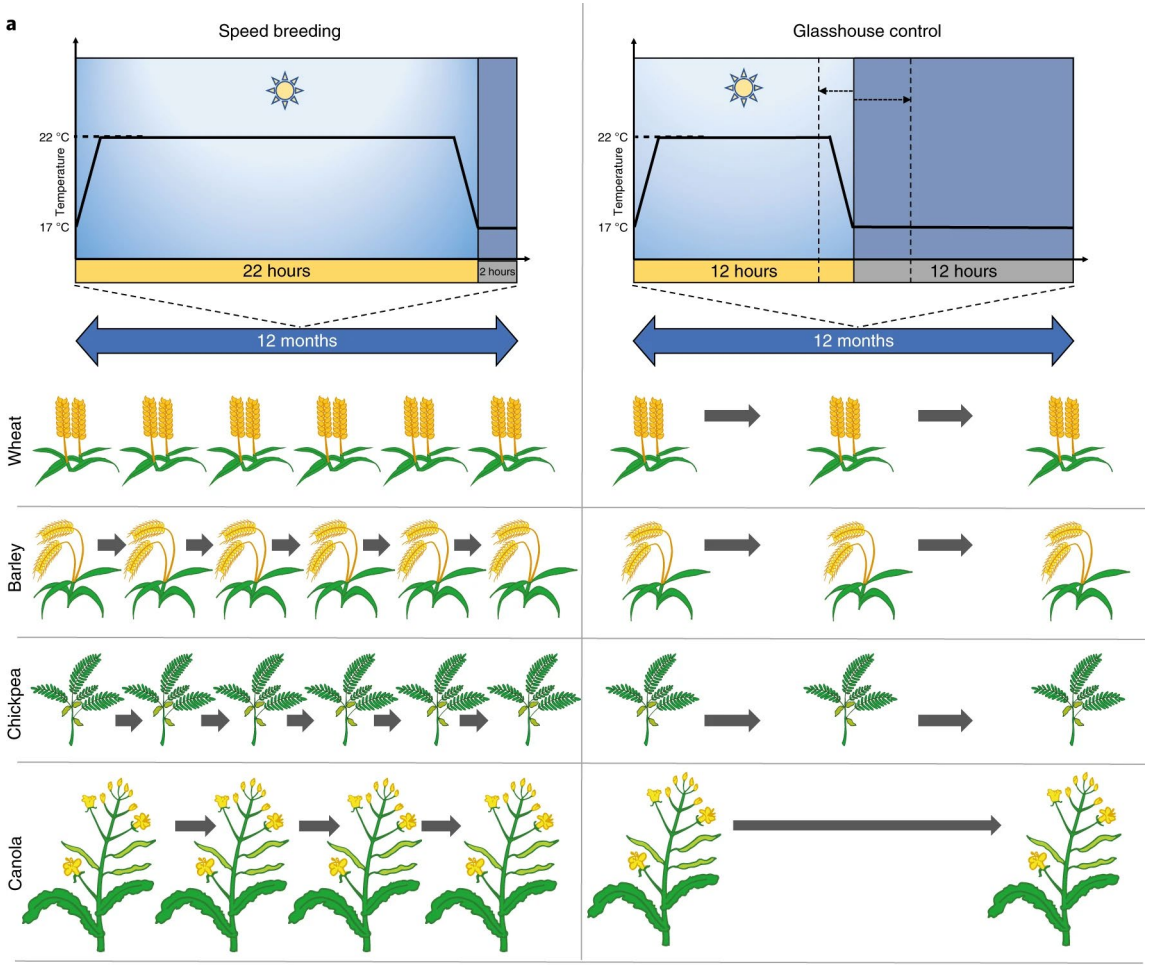
Genetic gain

$$\Delta G = \frac{ir\sigma_g}{L}$$

Cycle time

A key limiting factor for plant breeding: the long generation times.

Speed breeding: a breakthrough for rapid generation



Speed breeding of major crop plants

- 22/17 °C day/night temperature
- 65/85% day/night relative humidity
- 22 h / 2 h photoperiod
- ~500 μmol/m^2·s PPFD

spring type rapeseed:

~4 generations/year

Watson et al. 2018, 4:23-29

An ideal speed breeding system should be

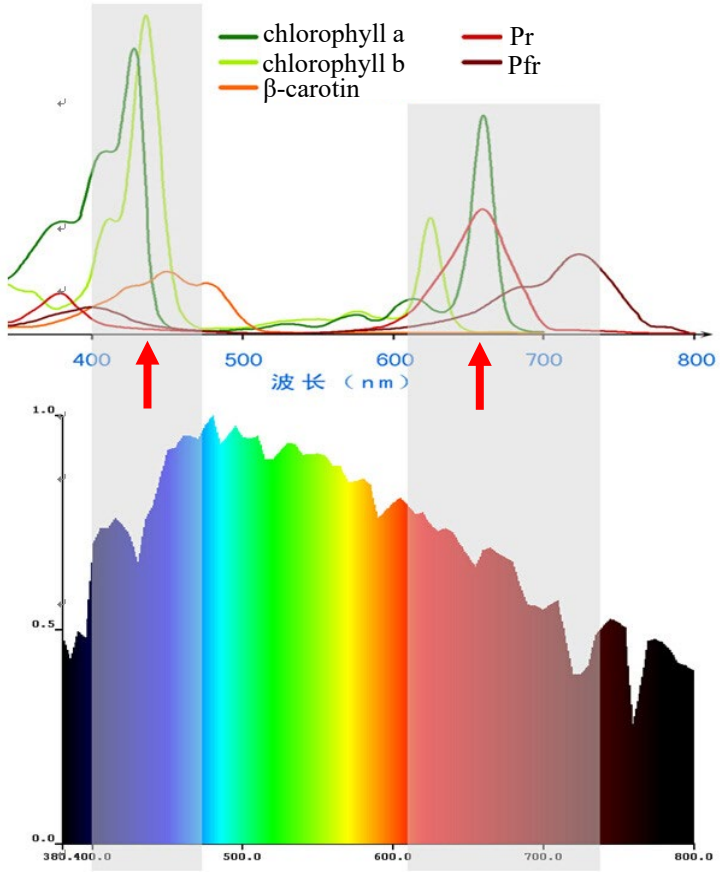
- Compatible for different ecotype
- Simple and efficient
- High-throughput
- Suitable for phenotypic investigation

- An optimum environment conditions (light)
- A fast vernalization method (for winter and semi-winter type)

Part II

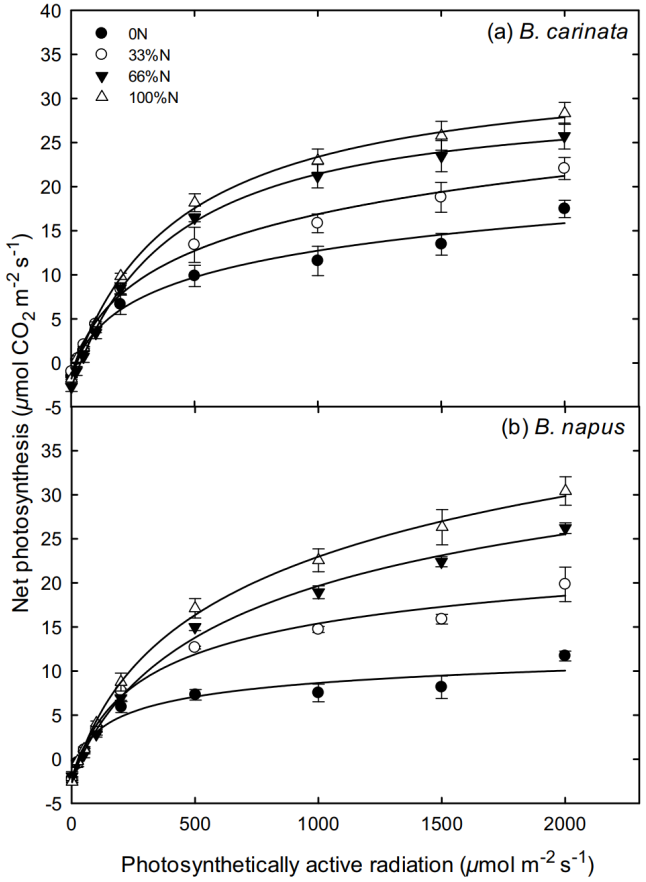
Comprehensive Speeding Breeding (CSB): a high throughput system for rapeseed

The photosynthesis characteristic of plants



Light absorption of plants and the spectrum of sun light

(Bantis et al. 2018)



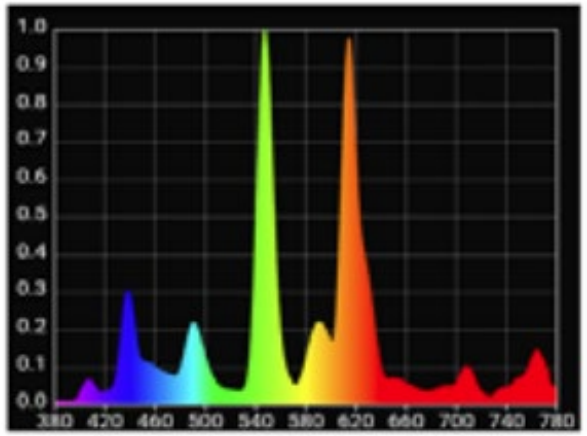
Net photosynthesis of *Brassica* as a function of photosynthetically active radiation

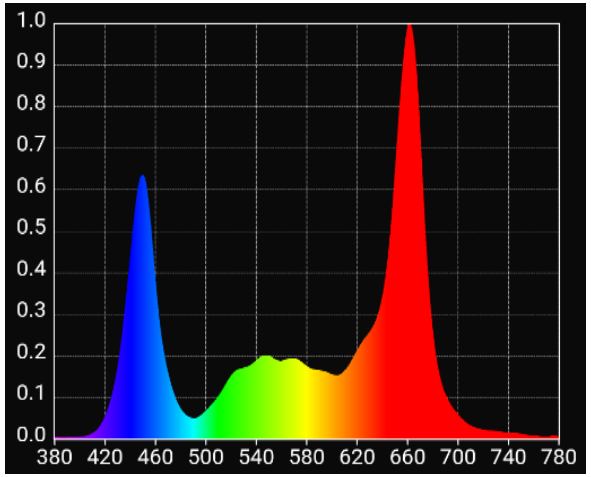
(Seepaul et al. 2016)

Rational design of the light spectrum and intensity is important in speed breeding system.

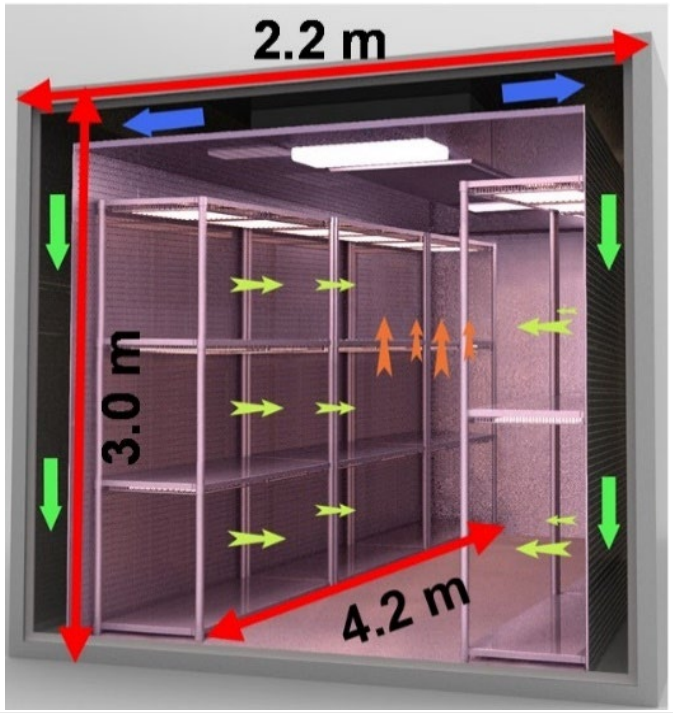
Optimized the SB system

Fluorescent lamp





Customized LED



10.8 m^2 growth area

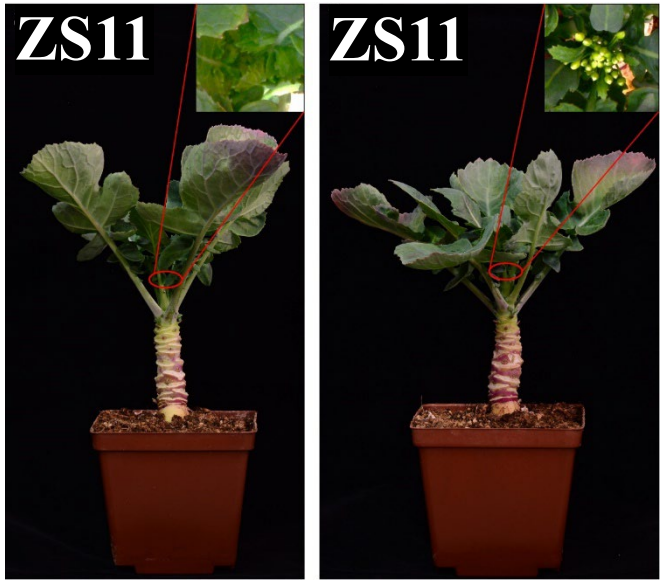
- 22 ℃ all day
- 22 h light +2 h dark photoperiod
- ~ **950 µmol/m^2·s** (10 cm below the LED)
- **R:G:B=6:2:2**

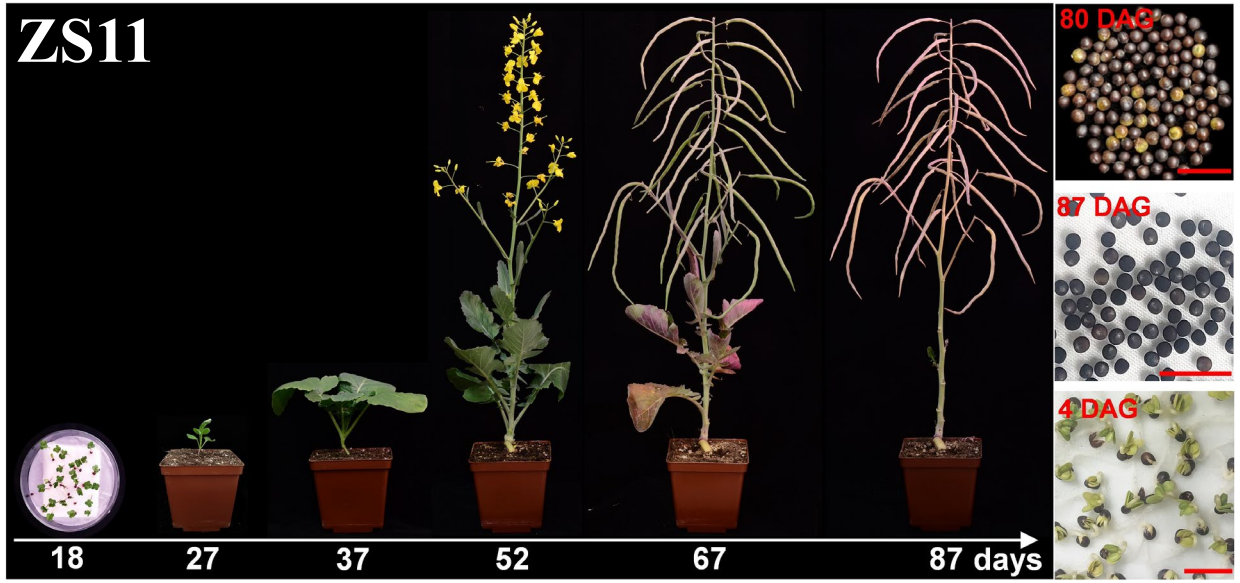
Rapeseed reactions to the new SB system

5.5 generations per year Westar (no vernalization)

40% in advance (113 to 67 days)

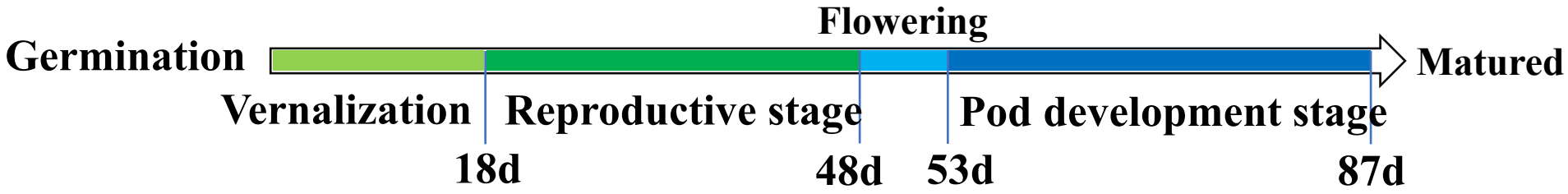
Rapeseed reactions to the new SB system





145 DAG without vernalization for semi-winter Zhongshuang11

4.5 generations per year with 17-d germinated seeds vernalization

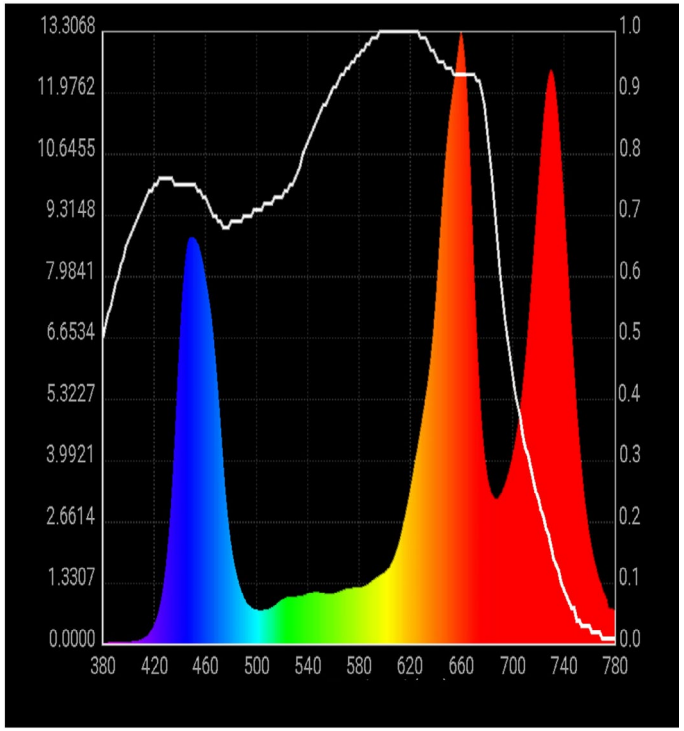


Rapeseed reactions to the new SB system

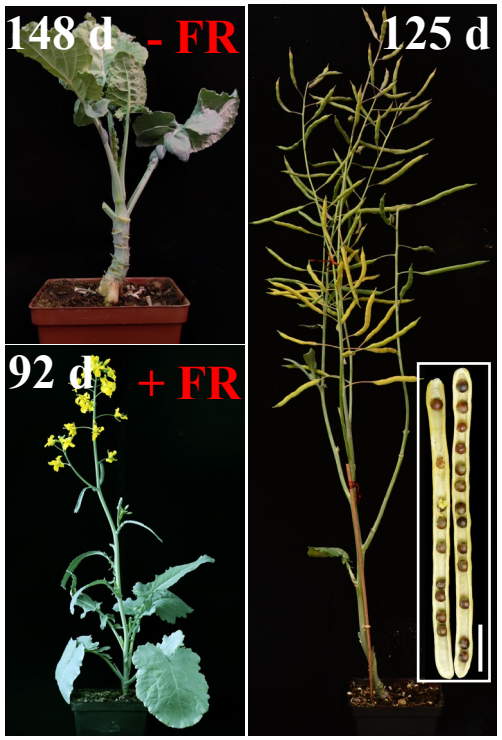
Despite 60 days of VGS, Darmor-*bzh* plants remained in the vegetative stage at 148 DAG

New SB system failed to work in winter rapeseed

Application of far-red light in speed breeding



Optimized spectrum



Darmor-*bzh*
(55 d vernalization)

Westar

ZS11

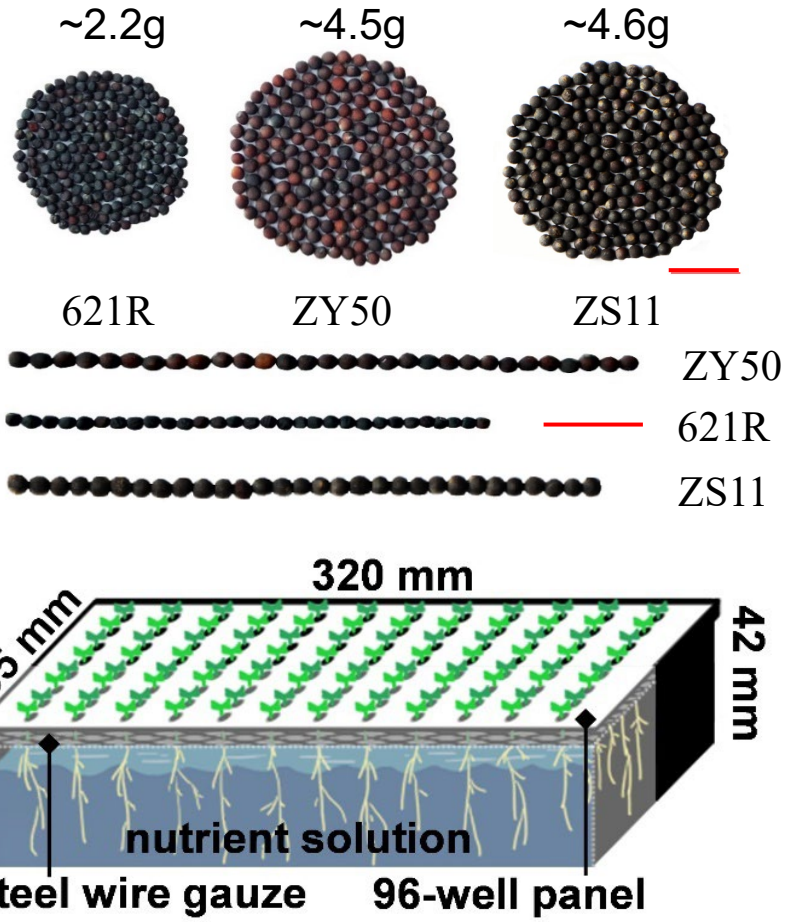
The life cycle of rapeseed could be further decreased with extra add of FR light

3-6.5 generations per year; for all types of rapeseed

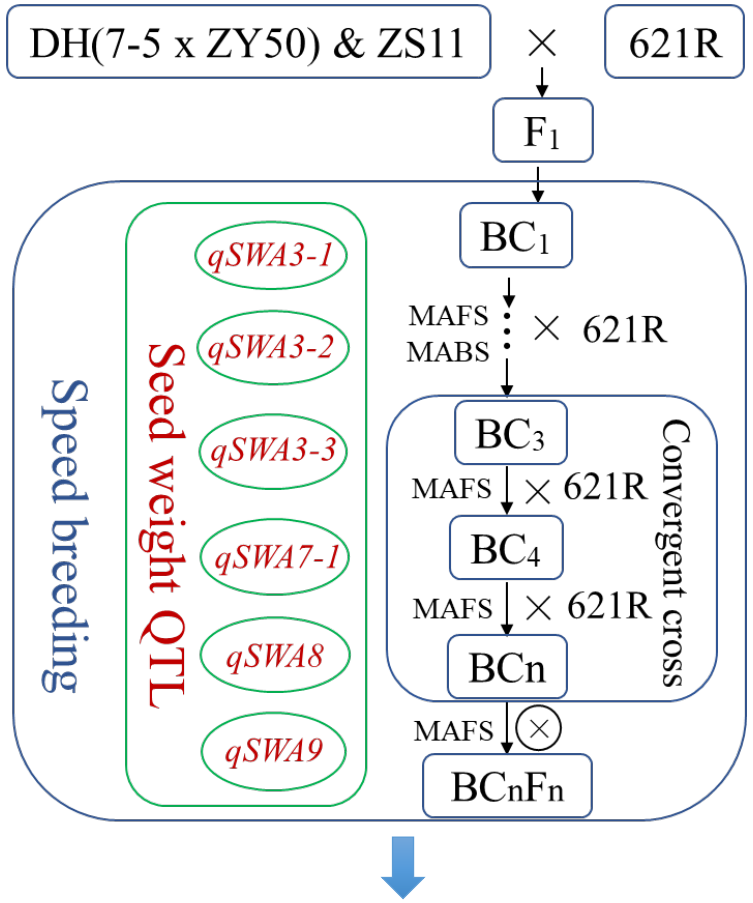
Part III

CSB is powerful for QTL pyramiding

QTL pyramiding to enhance TSW of an elite polima CMS restorer 621R



96-well seedling culture tray
(1000 plants/m^2)

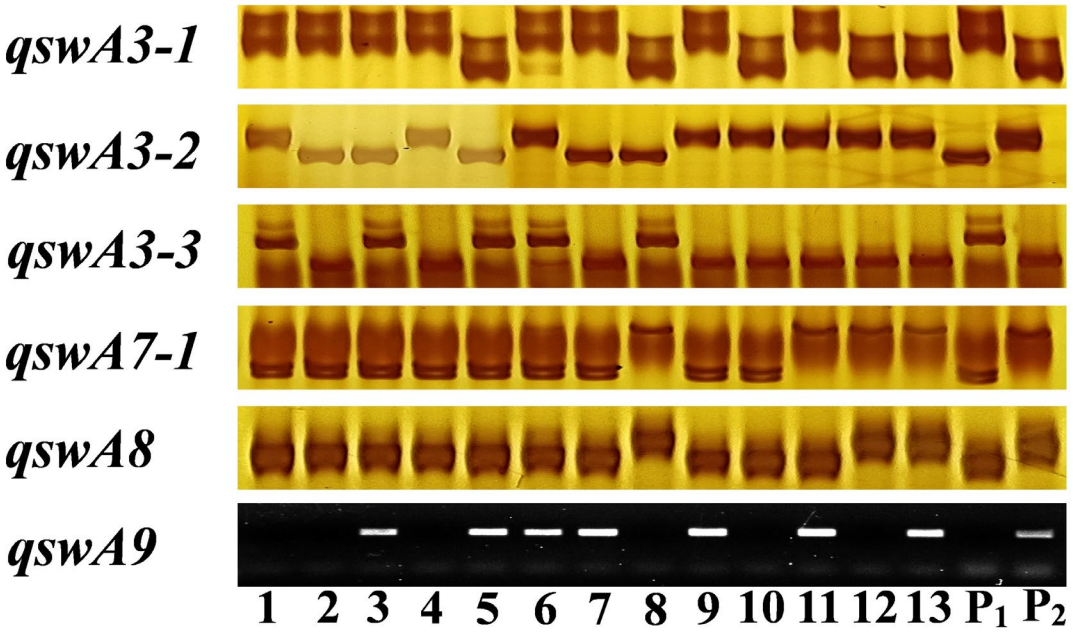


8 generations
in 23 months

Agronomic trait evaluation; F_1 hybrids

13 NILs with different QTL combinations were harvested

NILs
1. 621R$^{A3\text{-}2}$
2. 621R$^{A3\text{-}3}$
3. 621R^{A9}
4. 621R$^{A3\text{-}2/A3\text{-}3}$
5. 621R$^{A3\text{-}1/A9}$
6. 621R$^{A3\text{-}2/A9}$
7. 621R$^{A3\text{-}3/A9}$
8. 621R$^{A3\text{-}1/A7\text{-}1/A8}$
9. 621R$^{A3\text{-}2/A3\text{-}3/A9}$
10. 621R$^{A3\text{-}1/A3\text{-}2/A3\text{-}3}$
11. 621R$^{A3\text{-}2/A3\text{-}3/A7\text{-}1/A9}$
12. 621R$^{A3\text{-}1/A7\text{-}1/A8/A3\text{-}2/A3\text{-}3}$
13. 621R$^{A3\text{-}1/A7\text{-}1/A8/A3\text{-}2/A3\text{-}3/A9}$



Genotype of the BC_nF_5 lines

8 generations in 23 months

Improved lines quickly recovered the genetic backgrounds to 621R

A. 621R$^{A3\text{-}2}$

B. 621R$^{A3\text{-}3}$

C. 621R^{A9}

D. 621R$^{A3\text{-}2/A3\text{-}3}$

E. 621R$^{A3\text{-}1/A9}$

F. 621R$^{A3\text{-}2/A9}$

G. 621R$^{A3\text{-}3/A9}$

H. 621R$^{A3\text{-}1/A7\text{-}1/A8}$

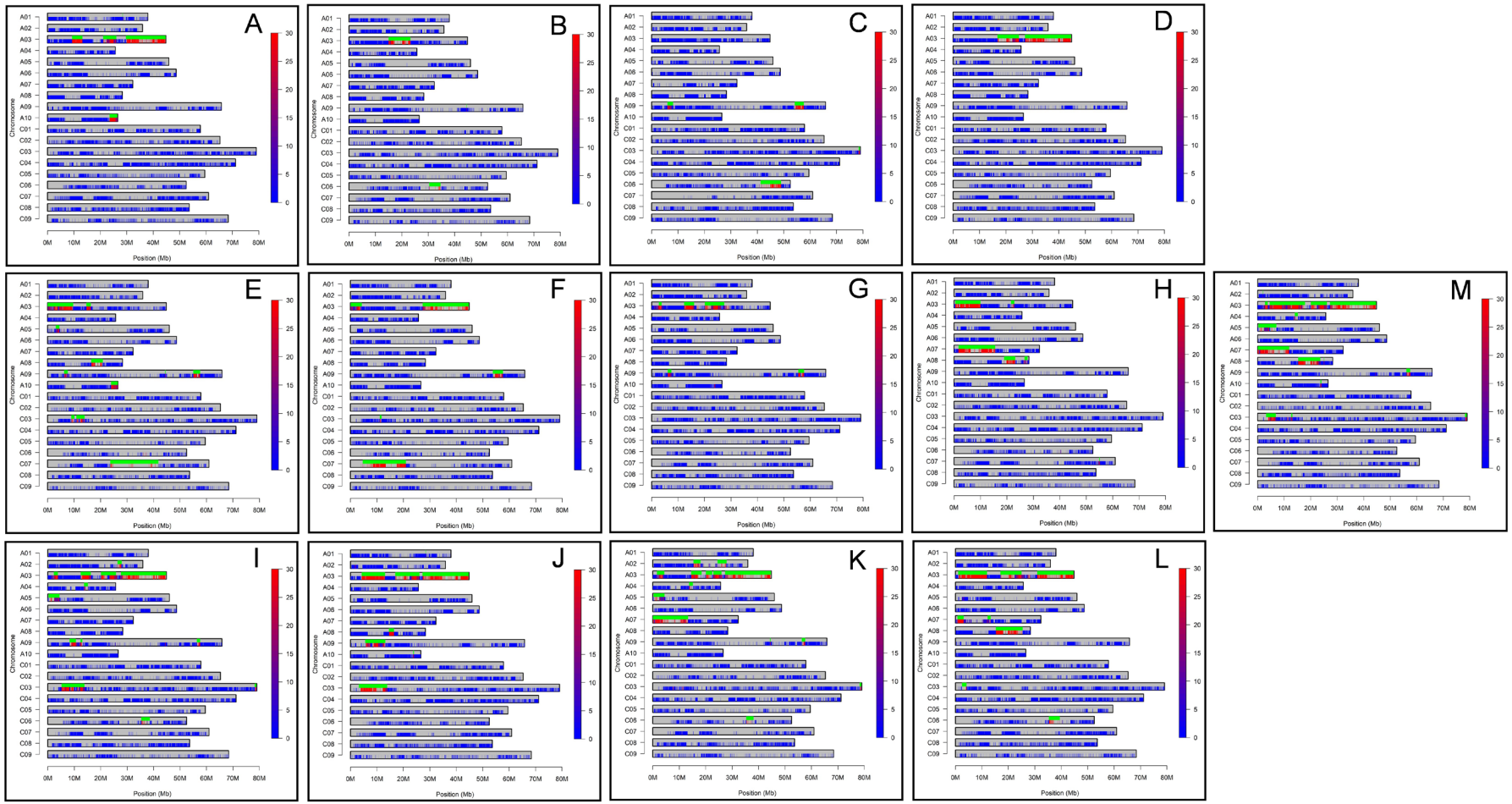
I. 621R$^{A3\text{-}2/A3\text{-}3/A9}$

J. 621R$^{A3\text{-}1/A3\text{-}2/A3\text{-}3}$

K. 621R$^{A3\text{-}2/A3\text{-}3/A7\text{-}1/A9}$

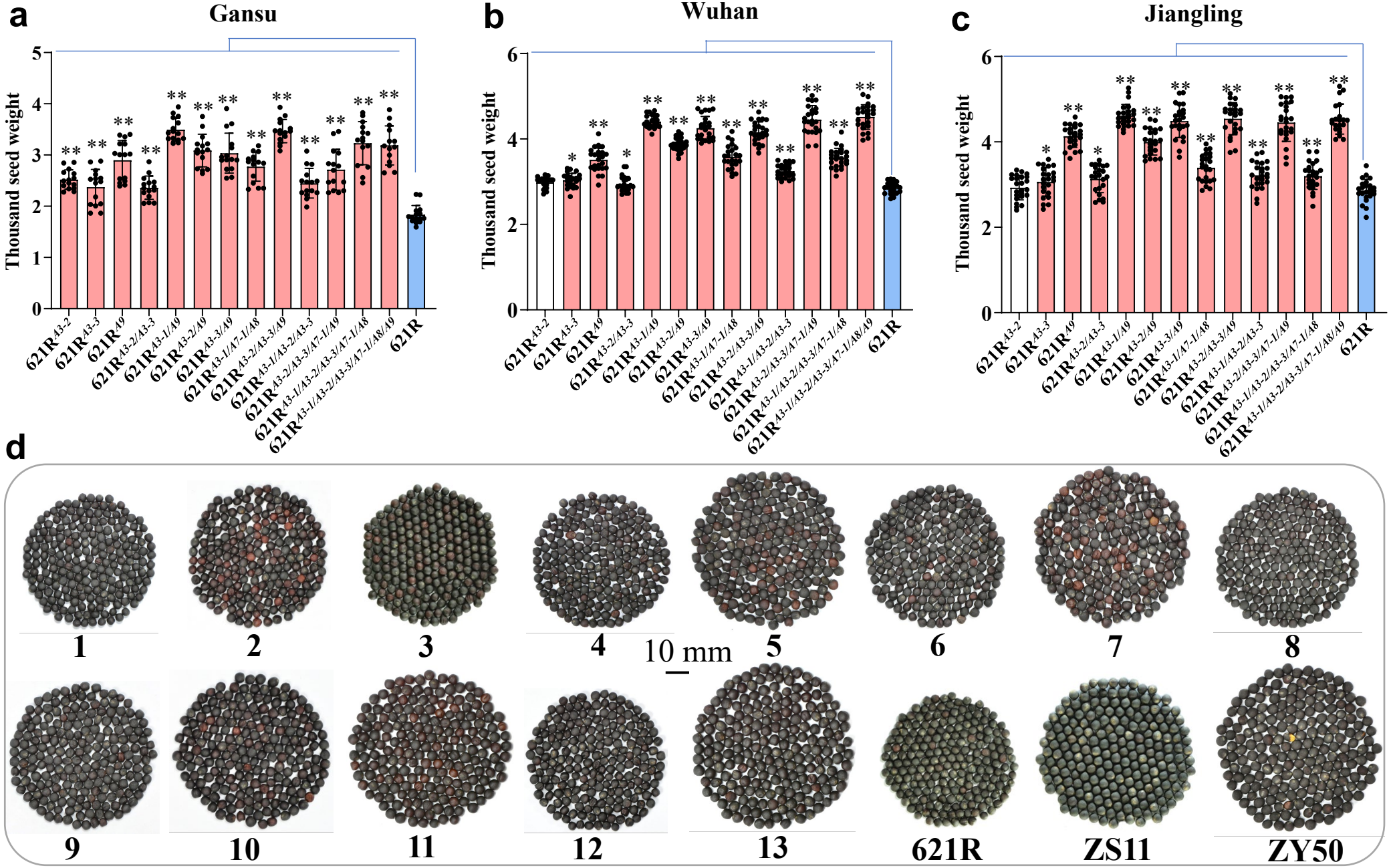
L. 621R$^{A3\text{-}1/A7\text{-}1/A8/A3\text{-}2/A3\text{-}3}$

M. 621R$^{A3\text{-}1/A7\text{-}1/A8/A3\text{-}2/A3\text{-}3/A9}$



The genome similarity are 97.17%, 98.27%, 98.56%, 97.34%, 95.02%, 95.57%, 97.72%, 96.69%, 93.69%, 94.07%, 93.79%, 93.88%, 91.79%, respectively.

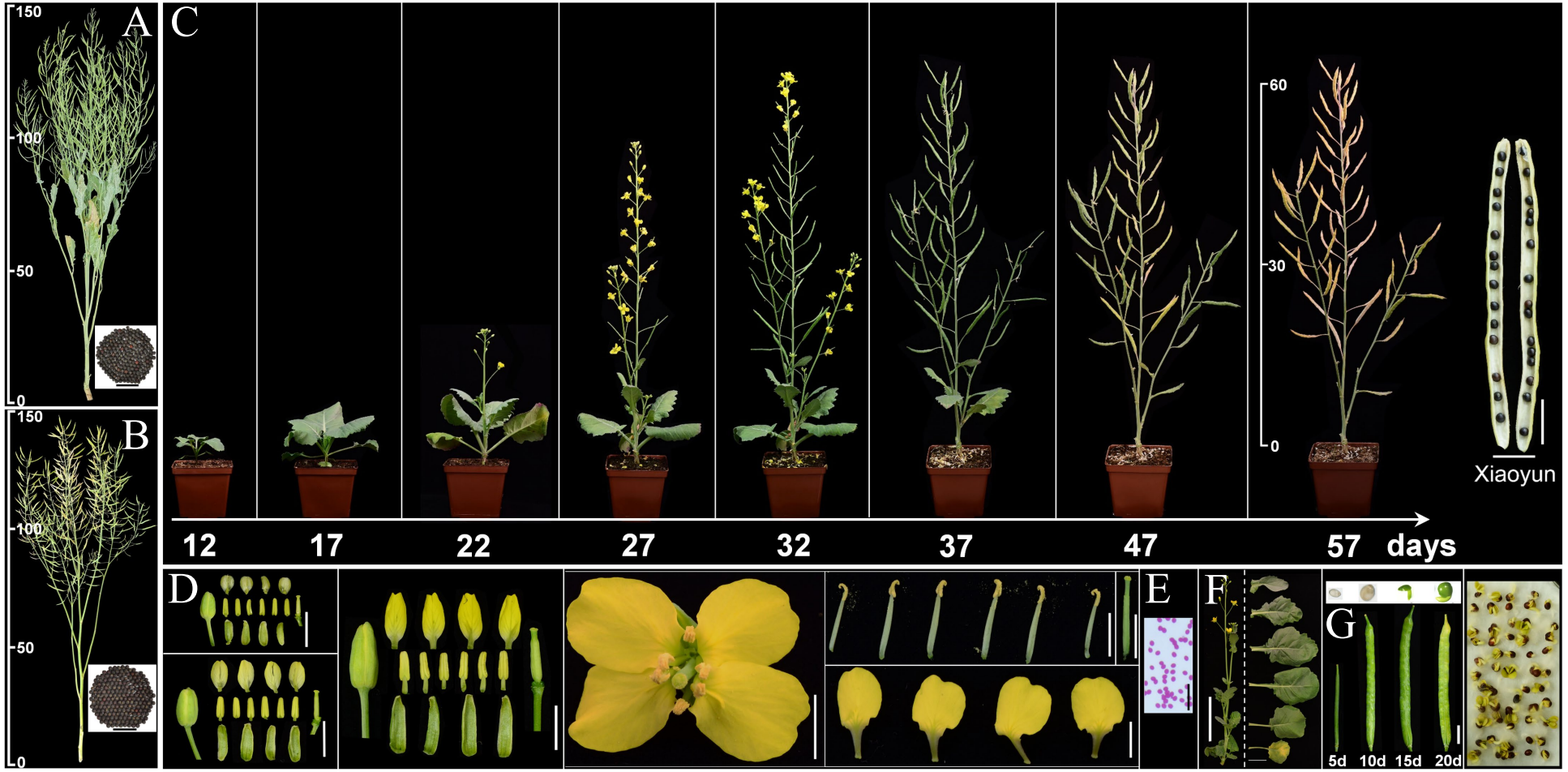
TSW of the 13 NILs was significantly increased



Part IV

CSB shows great potential for rapeseed functional genomics research

Screening of ideal genotype for rapeseed genomics research

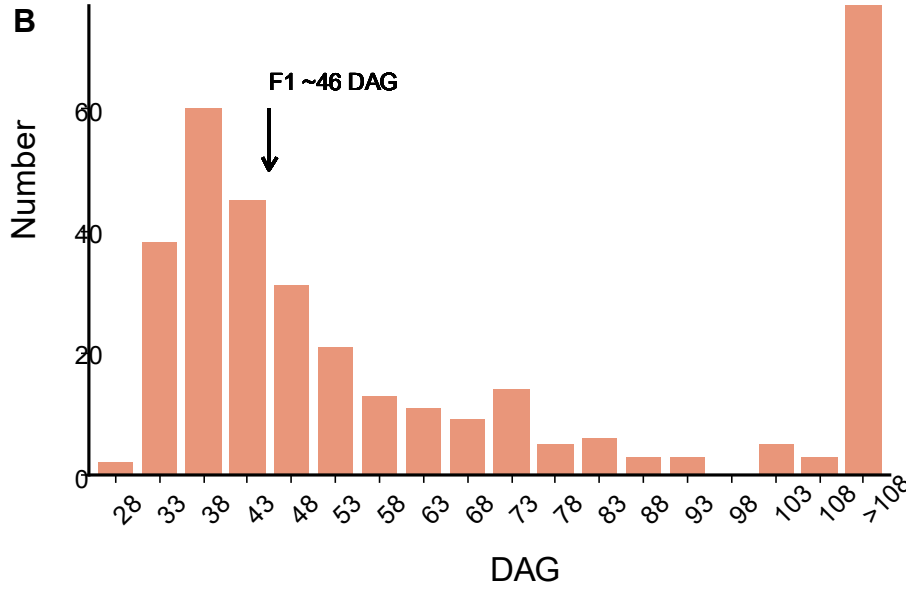


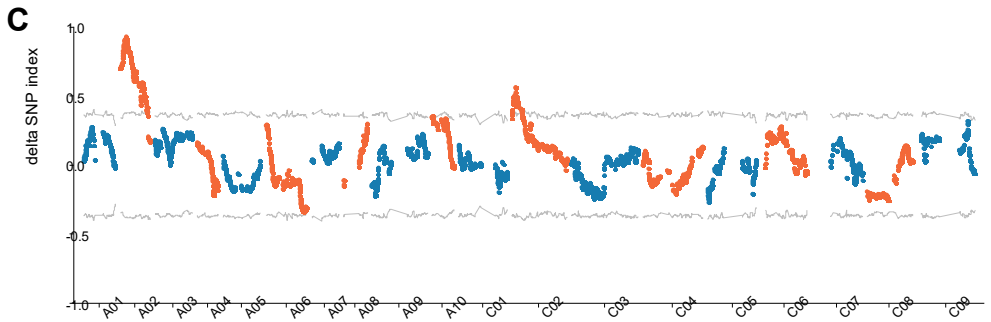
The phenotype of ‘Xiaoyun’ plants under modified speed breeding environment condition and field environment

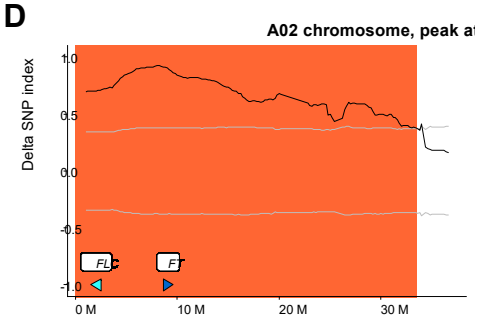
‘小芸 (Xiaoyun)’ shows extremely short life cycle (6.5 generations per year) without morphologic defect in SB conditions.

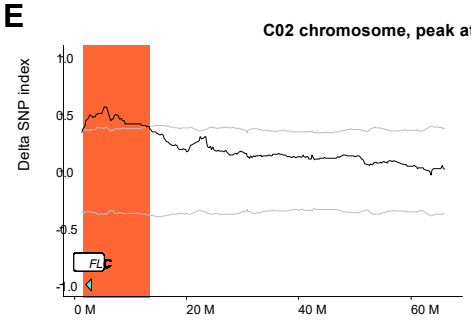
QTL-seq base on the phenotype from SB conditions

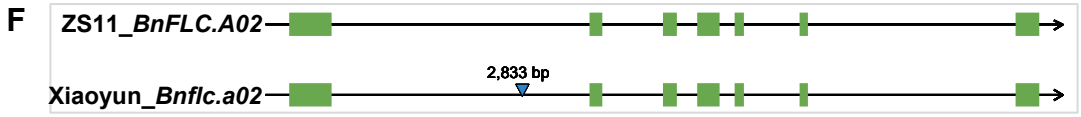
F_2 (Xiaoyun × ZS11)

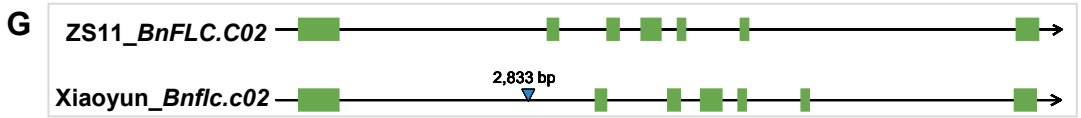


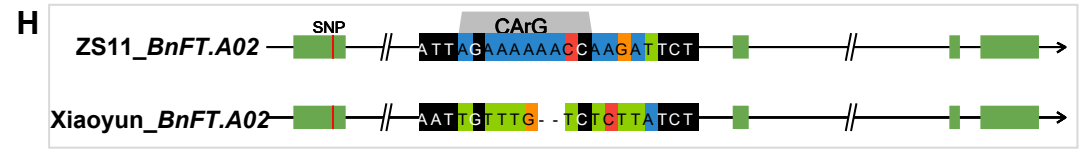




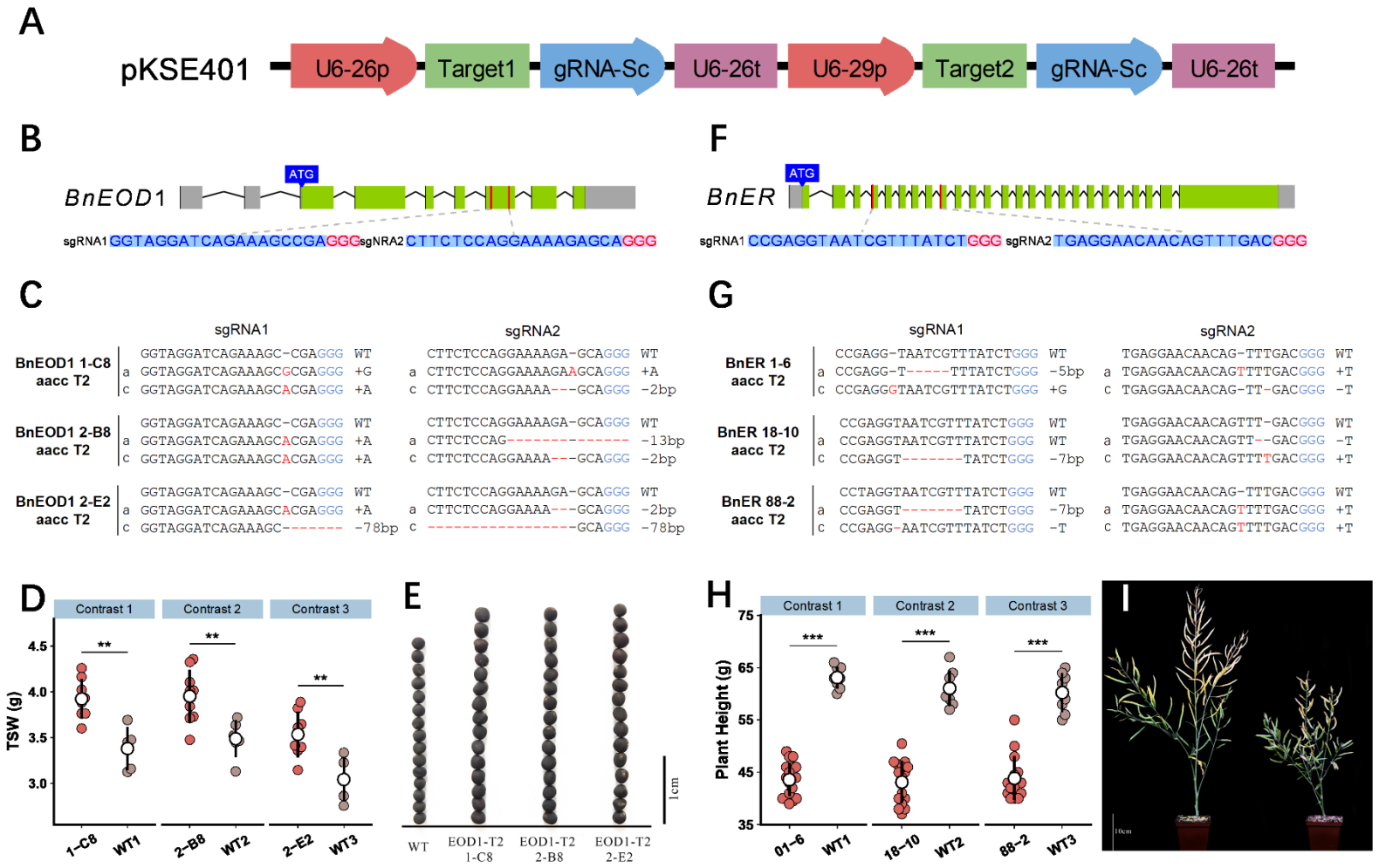








Quantitative trait analysis under SB



Xiaoyun's utility in quantitative trait research confirmed by CRISPR/Cas9 induced mutant of *BnEOD1* and *BnER*.

Acknowledgements

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