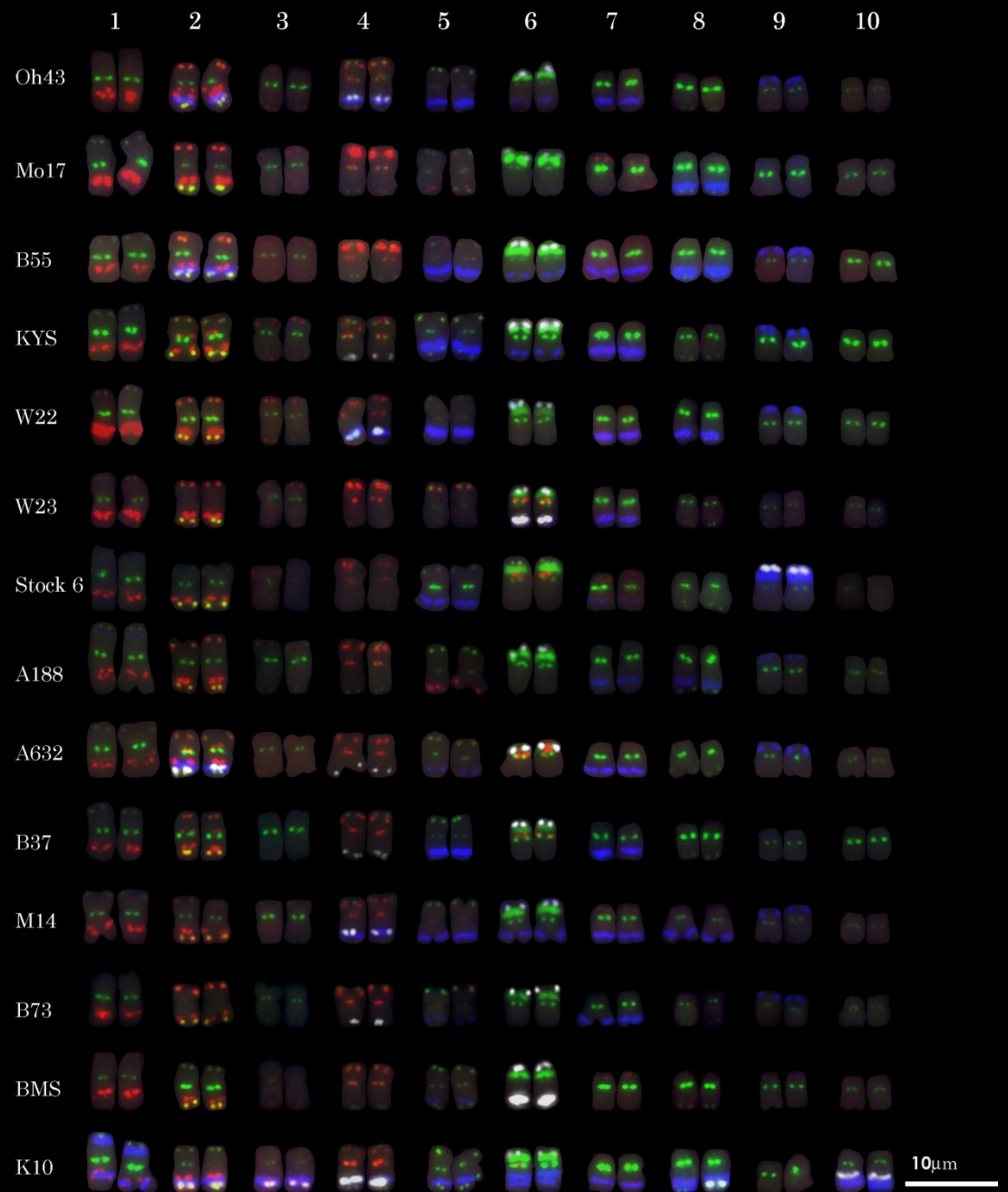


Construction and Applications Of Engineered Minichromosomes In Plants



Maize Chromosomes Of Common Inbred Lines

Oh43, Mo17,
B55, KYS, W22,
W23, Stock 6,
A188, A632, B37,
M14, B73, BMS,
K10



Engineered Minichromosome Construction

A major problem involving transgenic plants is the variability of expression of different insertion events.

Minichromosomes might serve as a platform for faithful expression of multiple transgenes on an independent chromosome.

Combined with haploid breeding, they could facilitate transfer of stacked transgenes among different varieties.

Artificial chromosomes could also be used to add whole biochemical pathways and new properties to plants.

An extra chromosome might allow plants to be used as a factory for the production of multiple foreign proteins or useful metabolites.

Telomere Truncation



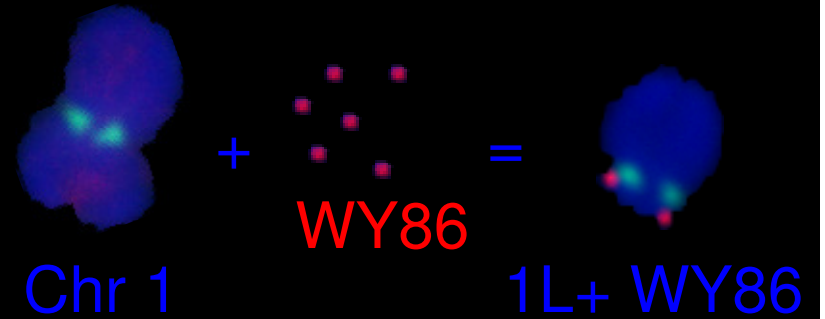
Transformation of telomere repeats (chromosome ends) will catalyze the formation of a telomere at an otherwise internal site.

When the chromosome is cut in this way, DNA sequences were also added that will allow further additions to the chromosome.

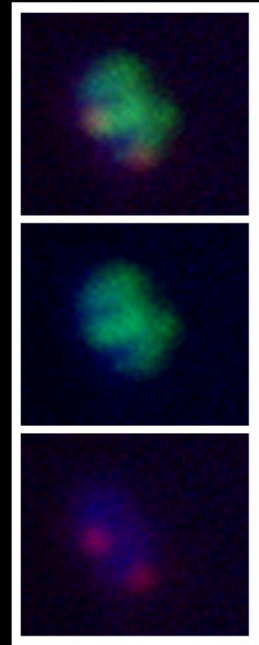
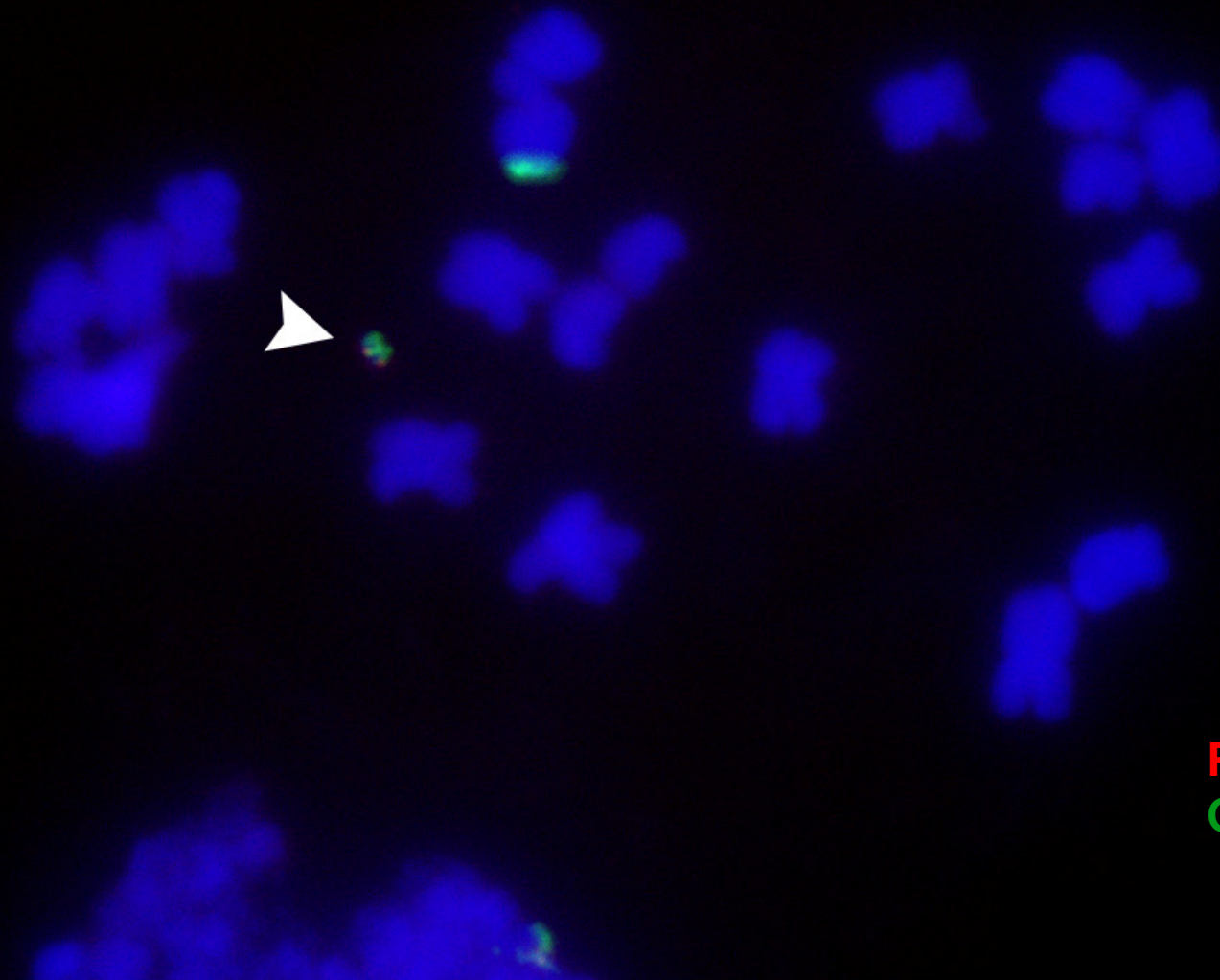
Agrobacterium transformation of a construct carrying many copies of the telomere sequence was performed in maize.

Truncations can be recognized because the transformation and cytological assay have no intervening haploid stage.

Minichromosome Construction

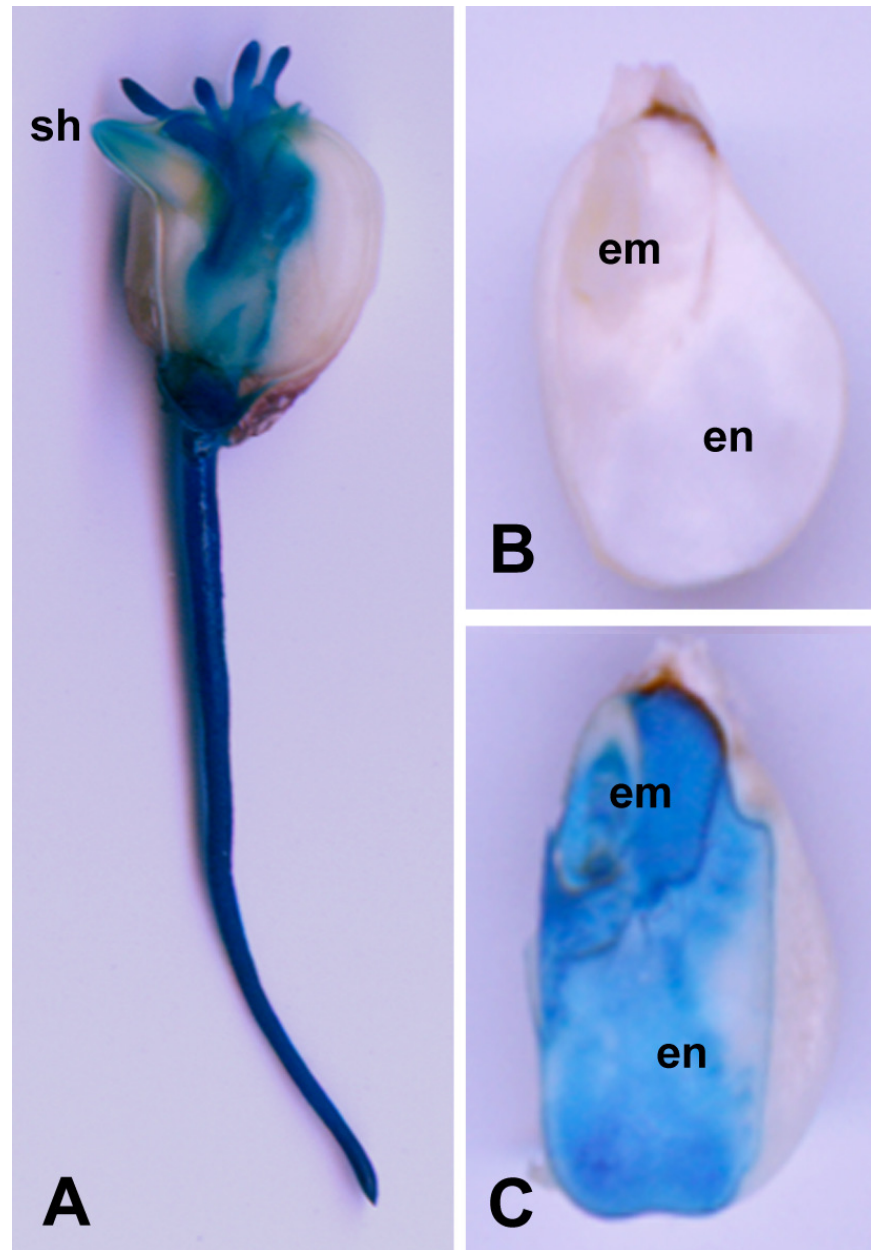


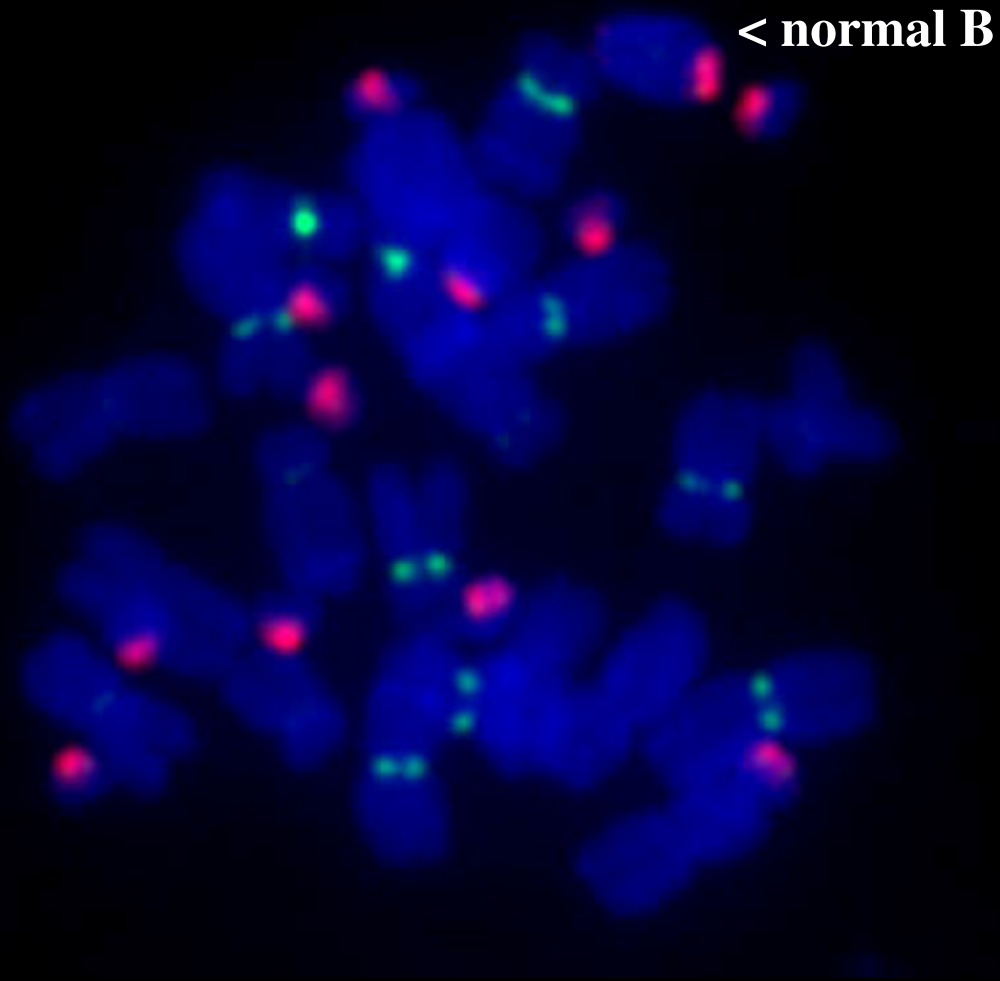
Smallest mini-chromosome



Red: Transgene
Green: B repeat

Reporter Gene Expression from a Minichromosome





B repeat = Red; CentC = Green

Dosage increase of a minichromosome to 11 copies

Plant artificial chromosome platforms

Can be initiated by telomere truncation.

Telomere truncation should easily work in most plants.

Can provide a platform for predictable gene expression.

Can add additional genes by site-specific recombination.

B (extra) chromosomes can serve as excellent target.

Highly deleted normal chromosomes can be recovered.

Potential for stacking a large number of transgenes.

Might be combined with 1x breeding to transfer to new lines easily.

Add new properties to plants.

Mass production of pharmaceuticals and useful metabolites.

Efforts in Arabidopsis, Brassica, barley, soybean, wheat and rice.

Engineering plants for food, fiber, feed, fuel and pharmaceuticals.



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