

MINICHROMOSOME TECHNOLOGY

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Genetic engineering is an important method for increasing crop quality and production while lowering labour and resource use in agriculture (Ceccarelli *et al.*, 1992). Traditionally, genetic modification has been accomplished by either Agrobacterium-mediated transformation (Opabode *et al.*, 2006) or direct transformation using a gene gun and particle bombardment (Altpeter *et al.*, 2005). Since these approaches allow for the insertion of single or few genes at random genomic locations and enable the simultaneous expression of several genes, they have many limitations; however, complex or combined traits cannot be transferred in a synchronised manner (Yu *et al.*, 2007b). For desired outcomes, these approaches are labour-intensive and time-consuming procedures that often necessitate highly qualified personal and substantial feedback. Furthermore, a large number of phenotypically abnormal plants are restored, and the host genome's utility is often disrupted.

Minichromosome technology offers a single solution for expressing and maintaining many transgenes in a single genome. Furthermore, plant artificial chromosomes or engineered minichromosomes may be a useful research method for deciphering chromosome structure and function. Since it is currently difficult to effectively insert massive repetitive DNA molecules into plant cells, minichromosomes, either naturally occurring or caused by irradiation, are an effective alternative for deciding minimum usable sizes of centromeres and constructing artificial chromosomes (Houben *et al.*, 2007 and Schubert *et al.*, 2007). Because of their ability to survive episomally, bear massive DNA inserts, and enable gene expression independent of the host genome, mammalian artificial minichromosomes have many potential biotechnological and therapeutic applications (Irvine *et al.*, 2005).

What is Minichromosome?

A minichromosome is a very small variant of a chromosome, which consists of thread-like linear or circular DNA and related proteins that contain genes and functions in the genetic material transmission process. Minichromosomes are plasmids that reproduce independently from *ori C*. (von Meyen burg *et al.*, 1979). They resemble their chromosomal counterparts in that they depend on functional *DnaA* and *DnaC* products, de novo protein synthesis, and RNA polymerase mediated transcription to initiate bi-directional replication (Messer *et al.*, 1996 and Weigel *et al.*, 1996). Transposable or repeated elements are also considered to be abundant in minichromosomes (Shiflett *et al.*, 2002). Radiation-induced breakages were the most common cause of minichromosome formation.

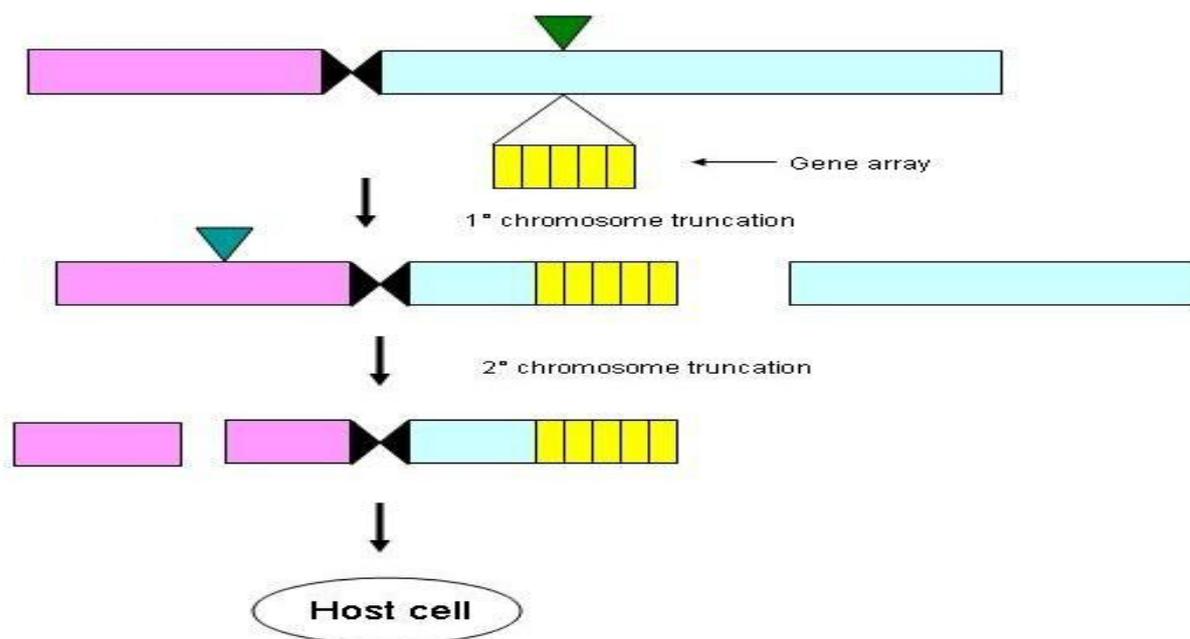


Fig 1: Mini chromosomes can be produced by telomere mediated chromosome truncation
(Aakash *et al.*, 2009)

Minichromosome in Plants

Previously, the role and application of minichromosomes were not well understood or recorded in the primary literature. Minichromosomes were later discovered to be very useful in understanding the fundamentals of chromosomal function and in plant genetic engineering (Birchler *et al.*, 2008; Houben *et al.*, 2008). Minichromosome technology has recently emerged as a powerful tool for improving crop plants.

Minichromosomes in Arabidopsis

A minichromosome was discovered in the telocentric line of *A. thaliana* using the Fluorescence In Situ Hybridization (FISH) method, and it was discovered to be from the short arm of chromosome number 4 (Murata *et al.*, 2006). This "mini4S" chromosome was estimated to be 7.5 Mb in size. Two additional minichromosomes (α , β and δ) have recently been found (Fig 2; Murata *et al.*, 2008). These two minichromosomes were discovered in an in-planta vacuum infiltration transgenic *Arabidopsis* plant.

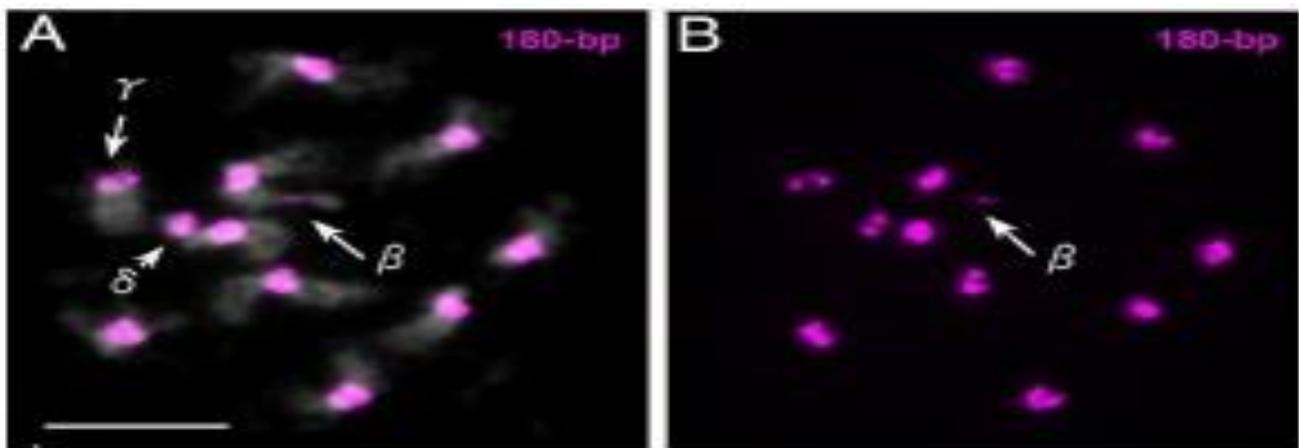


Fig 2: Cytological analysis of a G40 *Arabidopsis* cell containing minichromosomes α , β , and δ (Murata *et al.*, 2008).

Minichromosome in Maize

Maize minichromosomes have recently been generated by truncating the A and B chromosomes using telomere-mediated chromosome truncation (Fig. 3; Yu *et al.*, 2007a). Repeated backcrossing was used to move these minichromosomes to a diploid context to keep them stable. Although they produced A and B minichromosomes using this method, they were more interested in B chromosome-based minichromosomes because B chromosomes have a number of interesting properties (Kato *et al.*, 2005), including: (1). Unlike A chromosomes, truncation of B chromosomes would not induce developmental, or transmission complications, (2) Shape and the location of a B chromosome unique repeat in and around the centromeric region differentiate B chromosome derivatives, (3) Since there would be no residual endogenous genes to interfere with plant growth and transgene transmission, the size of mini-B chromosomes is unimportant.

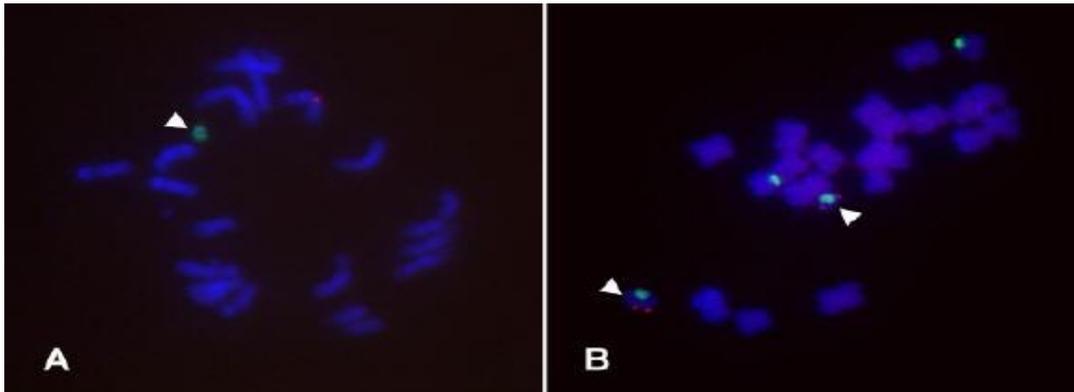


Fig 3: Mini chromosomes produced from maize B chromosome truncation, arrows denote mini chromosomes (Yu *et al.*, 2007a).

Conclusion

Any programme that involves the inheritance of several foreign genes as a unit may benefit from future innovations. Plants will also have whole biochemical pathways added to them in order to impart new properties or synthesise novel metabolites in large amounts. The quantity of mini-B chromosomes can be expanded to optimise the contribution from foreign genes on the minichromosome. Minichromosomes can be formed in most plant species due to the conserved telomere structure, allowing for a wide range of new applications in most agricultural crops.

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