**Construction and Characterization of a Deep-Coverage Bacterial Artificial Chromosome Library for Maize**

**2002**

Posted by: webmaster

Posted on: 2002/1/1 0:00:00

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**Abstract**

Modern cultivated maize (Zea mays L.) is one of the primary agronomic crops in the USA with an estimated genome size of 2500 megabases (Mb). To develop the resources for positional cloning and structural genomics in maize, we constructed a bacterial artificial chromosome (BAC) library for the inbred line B73 using the cloning enzyme Hind III. The library contains 247,680 clones (645,384-well plates). A random sampling of 697 clones indicated an average insert size of 136 kilobase (kb) (range = 42 to 379 kb) and 0.4% empty vectors. Screening the colony filters for chloroplast DNA content indicated an exceptionally low 0.18% contamination with chloroplast DNA. Thus, the library provides 13.5 haploid genome equivalents allowing >99% probability of recovering any specific sequence of interest. High-density filters were grided robotically using a Genetix Q-BOT (Hampshire, UK) in a 4 by 4 double-spotted array on 22.5-cm² filters. Partial screening (6× coverage) of the library with 20 single copy probes identified an average 7.1 positive signals per probe, with a range of 3 to 15 positive signals per probe. To evaluate the utility of the library for sequence tagged connector (STC) analysis, 768 BAC clones were end sequenced in both forward and reverse directions giving a total of 1415 successful reads. End sequences were queried against SWISS-PROT, Genbank NR, MIPS Arabidopsis, maize genomic sequence dbGSS, and maize cDNA database dbEST. Results in spreadsheet format from these searches is publicly available at the CUGI website (www.genome.clemson.edu/projects/stc/maize/ZMMBBb/).