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Toward Molecular Cytogenetical Characterizations in Cotton Genome

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Cotton is viewed as the most important cash crop in the world, and sustains the agricultural economies of many nations by providing a sustainable fiber product for the textile industry. Due to its global economic importance, many molecular tools are being developed. Florescent in situ hybridization (FISH), which allows DNA sequences to be mapped directly on chromosomes, is stressed as one of the most powerful techniques in plant molecular cytogenetics research. FISH has become an important tool and technology in plant genomics studies. It contributes many to genomics studies, mainly including: to confirm the correlation of the contigs with their respective linkage groups; to locate the contigs which contain a large number of repeats and are very difficult to be located using other technique(s); to confirm the connections between contigs; to identify or correct possible inversion(s) of genetic linkage map on chromosomes via orientation test; to identify possible chimeric contigs which locate on the end of chromosomes in genome sequencing; to delineate and characterize the information about gaps and be used as gap-closing tool in sequencing genome via DNA fiber FISH. FISH studies on rice, maize and wheat were successfully developed, but cotton lags behind the crops. Technologies of ISH (in situ hybridization) or FISH on cotton were established in the laboratory leaded by David Stelly in Texas A&M University. Since then, many contributions to cotton cytology, cytogenetics, and molecular biology have made by ISH or FISH. We recently developed cotton FISHs on meiotic pachytene chromosomes from Gossypium arboretum. Here we briefly summarize the technologies and primary studies in the diploid species. For preparing meiotic pachytene chromosome, the key steps were: (1) directly enzyme digestion to pollen mother cells rather than to buds, and (2) controlled spreading of the pachytene chromosomes using ice-cold acid-ethanol fixative. Following this way, it is easy to obtain well-differentiated bivalents with less cell damage and clear background and to identify all chromosomes respectively, because each chromosome has its distinct chromosome characteristics in the pachytene period. The 13 bivalents in pachytene period with various lengths exhibit a well-differentiated pattern of euchromatin and heterochromatin, which, along with chromosome length and centromere positions, permits the identification of all 13 bivalents. The results indicated that the technology developed by us can obtain well-differentiated pachytene chromosomes from cotton suitable for FISH. We do hope these can be well used recently on the ongoing genome sequencing species like G. raimondii for constructing its chromosomal physical map, further integrating the map with its genetic linkage map, and delineating and characterizing the information about possible gaps and even closing the gaps in the genome sequencing.