

4. Relationships between maize and human cytogenetics.

Considered in its broadest context cytogenetics has undergone a resurgence during the last ten years. In part, this renewed interest has been generated by advances in cell biology; likewise, the investigations in human cytogenetics have provided an additional stimulus. Particularly noteworthy from this latter area of interest are the workers, techniques and the problems. From the vantage point of an association with a medical school, there appears to be a paucity of discourse among groups of cytogeneticists. While professional isolation may in part account for this lack of interdigitation, certainly some of it arises merely from the ignorance that the other fellow exists. Intriguing cytogenetic problems are emerging amongst the mammalian and, in particular, the human cytogenetic areas: e.g. quantification of the variation in homologues; asynchronous replication of chromosome organelles; transmission of aneuploidy; the relatively high frequency of adjacent segregants from translocation heterozygotes (more often spoken of as the unbalanced carrier condition from a balanced carrier by the human cytogeneticist); as well as the more newsworthy association of chromosome abnormalities with disease (cause or effect?) and congenital abnormalities associated with aneuploidy.

It is to be hoped that there will develop a greater exchange between the largely research oriented cytogeneticists, employing predominantly corn and Drosophila technologies and the medically oriented, and undoubtedly motivated, human cytogeneticists. The case for understanding meiosis in man has been well put and considerable interest generated recently in attempting pachytene-diakinesis analysis, particularly during spermatogenesis. Without doubt, the extensive studies available from corn will provide a basis for human meiotic cytogenetic investigations.

Perhaps not so obvious is another area of joint interest. Since human cytogenetic studies have been almost exclusively a karyotype analysis of somatic material, the problem concerning the value of the karyotype as a predictor of meiotic behavior needs to be examined. Likewise, the accuracy of extrapolation from corn studies to humans can also be questioned. To provide evidence on these two questions, clearly mitotic karyotype procedures are needed in maize. Investigations of the mitotic karyotype and meiotic configurations in single plants or populations of plants could then be undertaken. Contributions from several laboratories, as noted in the last three Newsletters, have provided the necessary information and techniques so that it is now possible to undertake studies in maize employing a mitotic karyotype from root tips and the meiotic analysis of the same plant.

It is the purpose of this communication to bring these features to the attention of maize geneticists and to encourage this approach to specific problems in maize cytogenetics. Already we are impressed in our laboratory with several features of the mitotic karyotype-meiotic analysis protocol. Noteworthy at this time are: (1) the variability between homologues; (2) alterations in mitotic-meiotic arm ratios; (3) the use of chromosomal aberrations for experimental studies.

The availability of the large number of structural rearrangements and aneuploids in maize suggests that specific experimental procedures can be applied to problems of somatic cell cytogenetics in corn once the specific evidence on the relationship between the corn system and the human system has been presented. It can be anticipated that there could be a broad application of such corn studies. Not beyond a reasonable probability is

the promise of synthesizing a maize system to mimic a human chromosomal abnormality such that data would be available from progeny testing of maize prior to the attainment of reproductive age by the carrier of the abnormality.

We have been developing during the last 2½ years in this laboratory a catalogue of the karyotypes of various standard stocks--both hybrid materials and genetic tester stocks. We would be particularly interested to receive from other laboratories stocks containing known structural abnormalities not already available through the Co-op or stocks possessing chromosomes which appear cytologically "abnormal."

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5. Root tip squash technique.

The following is the procedure of root tip squash technique presently used in our laboratory.

1. Pretreat the excised root tips in 0.002 M 8-hydroxyquinoline for three hours. Root tips can be obtained by germinating seeds in petri dishes or by growing plants in pots. In both cases, we usually keep the seedlings at 28-30°C.
2. Fix in a mixture of one part of acetic acid and three parts of absolute alcohol overnight.
3. Wash with 70% alcohol and store in the same fluid in a refrigerator until needed.
4. Hydrolyze in N HCl at 60°C for 8 minutes.
5. Rinse in distilled water for three changes, 2-3 minutes each change.
6. Stain in leuco-basic fuchsin for 1-2 hours.
7. Treat with 5% pectinase for two hours.
8. Put in 45% acetic acid for about 10 minutes before squashing to clear the stain in cytoplasm.
9. Squash the meristematic regions in 45% acetic acid.

We found this technique quite satisfactory for the somatic chromosomes of maize. The ten pairs of chromosomes can be identified without much difficulty by their relative lengths, arm ratios and presence or absence of a satellite at metaphase. The eu- and heterochromatic regions and even some knobs can be differentially stained at prophase. Using this technique, we are studying the karyotypes of several diploid strains and translocation stocks and identifying the extra chromosomes of different trisomics.

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