

Microarray Technology Shown to Improve Diagnosis

Dr. Angie Dawson received an inaugural DSM grant in 2008 for her research project involving the use of microarray technology in the analysis of pediatric leukemia. This project was done in collaboration with Drs Rochelle Yanofsky and Sabine Mai from CancerCare Manitoba.

For over 30 years, the first-line test for identifying genetic disorders in individuals with developmental disabilities, multiple congenital abnormalities or cancers, such as leukemias and tumours, has been an assay in which the technologist looks at chromosomes under a microscope, looking for numerical and structural rearrangements. This technique is referred to as karyotyping.

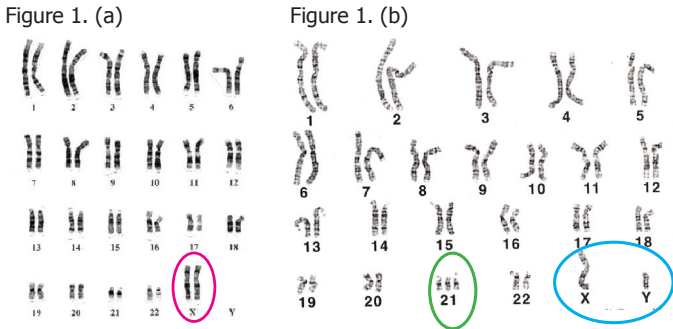


Figure 1. (a) normal female karyotype: 46,XX; (b) Down syndrome male with trisomy 21: 47,XY,+21.

Conventional karyotyping has a resolution limit of 5 Mb (5 million base pairs) of DNA and is the best choice for testing for genetic disorders, such as Down syndrome, that are caused by gross chromosome abnormalities. Examples of a karyotype are shown in Figure 1. Humans have 46 chromosomes, which are arranged in 23

pairs, numbered 1-22, with the 23rd pair being the sex chromosomes. A female has two X chromosomes (Fig. 1a; XX in pink) and a male has one X and one Y chromosome (Fig.1b; XY in blue). Down syndrome is the most common cause of mental handicap in humans and is caused by three copies of chromosome 21, resulting in 47 chromosomes (Fig. 1b; +21 in green).

Most developmental disorders arise from more subtle genetic alterations that are much smaller than 5 Mb. This is where microarrays come in. About five years ago, a new chromosome technology called microarray, which provides a much higher molecular resolution of chromosomes, became available. Microarrays can detect smaller changes in DNA content, such as deletions and duplications that are 500 Kb (500,000 base pairs), or less. However, microarrays cannot detect rearrangements that do not contain quantitative changes in the amount DNA, such as translocations or inversions. Therefore, continued routine conventional cytogenetics remains essential.

An example of a microarray is shown in Figure 2. A patient referred to genetics for a clinical indication of developmental delay was initially reported as having a normal conventional karyotype. Microarray analysis showed a small, 200 Kb (200,000 base pair) deletion (red) of chromosome band 22q11.22. This deletion is much too small to be seen down the microscope. All other chromosomes were normal by microarray analysis.

Many chromosome rearrangements have also been described in patients with acute lymphoblastic leukemia (ALL) and their association with specific clinical, biological, and prognostic features is well defined. Treatment failures can occur unpredictably in ALL patients with a favorable prognosis and 'good' cytogenetics at diagnosis.

In the current study, microarray technology was applied to nine retrospective cases of pediatric ALL with either initial high risk features or at least one relapse. Our hypothesis was that the presence of undetected chromosome DNA deletions/duplications in these patients would account for their poorer prognosis. In order to determine if the abnormalities, as interpreted by conventional cytogenetics, were consistent with the microarray results, the conventional karyotype of the pediatric ALL cases was compared to the microarray results. Of the nine cases examined, two cytogenetic ALL karyotypes were consistent with microarray data: one of the two showed a cytogenetic translocation that was not detected by microarray. Microarray identified chromosome losses and gains in the remaining seven cases, which were not detected by conventional karyotype. In addition, despite best efforts, the cytogenetic interpretation of the origin of several marker chromosomes was not consistent with the microarray results and required re-analysis of the chromosome rearrangements. The presence of these previously undetected abnormalities may help explain the poorer outcome and more difficult course of these patients.

It is our conclusion that the complementary use of microarray and conventional cytogenetics would allow for more sensitive and accurate analysis of the underlying genetic profile with concomitant improvement in prognosis and treatment, not only for pediatric ALL, but for all neoplastic and constitutional disorders in general.

Publications:

Dawson AJ. 2009. Utilizing CMDX's TOP (technical only program) for Implementing Array CGH Services in Canada. Invited speaker, Comprehensive Array Symposium and Exhibition (CASE). Combimatrix Molecular Diagnostics (CMDx), Irvine, CA.
 Dawson AJ, Yanofsky R, Mai S. 2009. Application of array comparative hybridization to the cytogenetic analysis of pediatric acute lymphoblastic leukemia. Presented at the 59th Annual Meeting of the American Society of Human Genetics, Honolulu, Hawaii.
 Dawson AJ, Yanofsky R, Vallente R, Bal S, Schroedter I, Liang L, Mai S. 2010. Microarrays and the Cytogenetic Analysis of Leukemias. Accepted, Current Oncology.

Figure 2 Chromosome 22

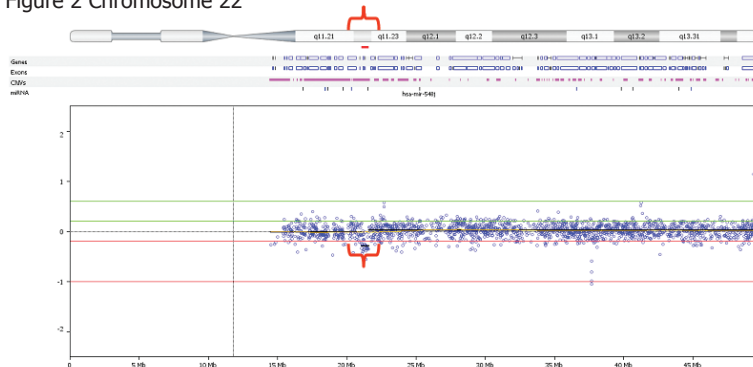


Figure 2. A microarray scan of chromosome 22 showing a 200 Kb deletion (red) in a patient with developmental delay. Each blue dot in the microarray represents a piece of DNA from chromosome 22.