Case 1: Clinical Report and Molecular Cytogenetic Findings

- 3½ month old female previously seen in the newborn period for multiple congenital anomalies.
- Patient findings are strongly suggestive of CHARGE Association including congenital heart defects, coloboma and growth retardation.
- Family history of a full brother who was stillborn with unknown etiology.
- High-resolution G-band studies revealed a 46,XX karyotype.
- Subtelomeric DNA Copy was normal.
- FISH studies utilizing a homebrew locus specific DNA probes revealed a deletion of the CREBBP gene at 8q22. Deletions of this gene have been associated with CHARGE.
- Microarray analysis utilizing the Spectral Genomics™ Constitutional Human BAC Array was normal.
- Microarray analysis with the Spectral Genomics™ 2600 BAC 1Mb Array confirmed a deletion at 8q12.2 but also revealed a duplication at 1q44 and 1p36.32-33 (Figure 3a). A deletion at 8p14.3 and duplications at 14q12 and 22q13.33 were also present.
- FISH studies utilizing homebrew loci specific DNA probes derived from the same clones found to be duplicated at 1q44 (RP11-407H12 and RP11-438F14) in the microarray study were positive for a duplication in approximately 1/3 of the interphase cells scored (Figure 2a and 2b).
- Microarray and FISH studies of the parental chromosomes revealed the inheritance pattern of the suspected LCVs.
- Further investigation revealed that the clones duplicated at 1q44 (RP11-407H12 and RP11-438F14) and 1q42 (RP11-125AS) have been previously reported as polymorphic clones located within a LCV region. The clone showing gain at 8p14.3 (RP11-88L18) to the best of our knowledge, however, is not within a reported LCV.

Large-scale Copy-number Variations (LCV): What do we know so far?

- What are they? LCVs are large segments of DNA, several to hundreds of kilobases in size, that vary severalfold in copy number between phenotypically normal individuals.
- How are they detected? LCVs manifest themselves as duplication/losses in DNA microarray studies and/or deletions in FISH studies. They may also be seen to a lesser extent as duplications in interphase FISH studies.
- How many are there? Estimates of greater than 200 LCVs are present in the human population. This is based on limited population studies and may be an underestimate.
- How common are they? On average, two individuals vary by 11 or more LCV’s, however less than 50% of the LCV’s currently described were identified in more than 1 individual.
- Where are they located? Greater than 1/3 of the polymorphic clones may overlap known coding regions and may encompass one or more complete genes. Several LCV’s have been identified near loci associated with cancer or genetic syndromes and are frequently in regions susceptible to rearrangement.

LCV’s: What does it mean when they are observed in affected individuals?

Although LCV’s are considered normal genomic variants, the possible phenotypic consequences of all LCV’s has yet to be determined.

- Some LCV’s in deletion may uncover recessive mutations resulting in phenotypic changes.
- Some LCV’s may have yet to manifest themselves in age-related susceptibilities.
- Some LCV’s may lead to further genetic alterations within adjacent regions.

Conclusions about associations between LCV’s and phenotypic abnormalities should be approached cautiously.

- An aberration in a LCV region should be considered as a possibility for the patient’s malformation only if it has been observed in other patients with similar phenotypes or is associated with regions containing genes which may have contributed to the abnormal phenotype.
- Aberrations observed by either DNA microarray or subtelomeric FISH studies that have not been associated with a specific phenotype in other case studies or cannot be tied to a possible causative gene within that region may represent a previously unreported LCV region.
- Parental studies are necessary to help classify all possible LCV regions.

To achieve optimal utilization of molecular cytogenetic techniques and to enhance our basic understanding of LCV’s it is of great importance to report cryptic aberrations thought to be genomic polymorphisms and this must be accumulated in a national database to allow clinical laboratories to accurately determine whether duplications or deletions, by these methods are clinically relevant.