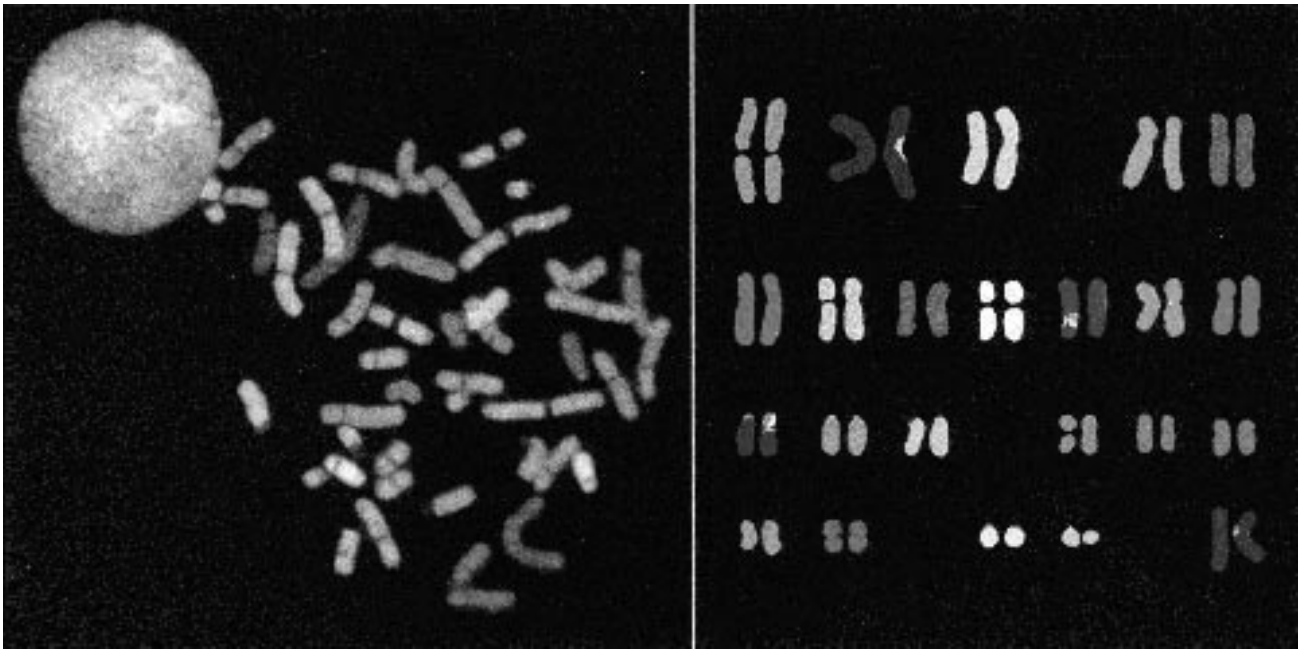
Molecular Biology, VI



Spectral karyotyping of human chromosomes after the simultaneous hybridization of 24 combinationally labeled chromosome painting probes. Left panel: presentation of display colors. Right panel: karyotype of the classification-colored metaphase chromosomes

Metaphase Preparation of Normal Human Chromosomes With Spectral Karyotyping

Cancer cytogenetics and gender diagnosis routinely include a light microscopic analysis of human chromosomal number and structure. Beyond this routine karyotyping, fluorescence in situ hybridization has been applied to increase the sensitivity, specificity, and resolution of detection of chromosome aberrations. Now a spectral imaging approach has been developed to allow the use of multiple fluorescent probes that overlap spectrally to characterize human chromosomes. Spectral karyotyping (SKY) allows the simultaneous visualization of all human chromosomes with each chromosome in a different color. SKY is based on the hybridization of 24 human chromosome painting probes labeled with different fluorochromes or fluorochrome combinations. The process of image creation and acquisition is based on a combination of conventional fluorescence microscopy, charge-coupled device (CCD) imaging, and Fourier spectroscopy. In contrast to the application of fluorescent specimen using optical-filter-based approaches, spectral imaging allows the measurement of the spectrum of the emitted light at all image points simultaneously. This allows the creation of probes that color the genetic material from each chromosome distinctively. Thus, SKY can be applied to study constitutional chromosomal aberrations in the prenatal or postnatal cytogenetic laboratory, and it provides a useful tool with which to comprehensively reconstruct complex chromosomal aberrations in leukemia and solid tumors. Moreover, this technique of karyotype analysis based on color information has been automated. The method has been described in detail by Schröck et al. (Science 1996; 273:494–497).

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