characteristic translocations is also aided by molecular techniques, in part because chromosome preparations are often difficult to obtain from solid tumors. For example, many sarcomas of childhood, so-called round blue cell tumors (Chapter 10), can be difficult to distinguish from each other on the basis of morphology. However, the presence of the characteristic (11;22)(q24;q12) translocation, established by PCR, in one of these tumors confirms the diagnosis of Ewing sarcoma. Another diagnostic platform that is finding increasing use is **DNA microarrays,** either tiling arrays, which cover the entire human genome, or single-nucleotide polymorphism arrays (SNP chips), which allow high-resolution mapping of copy number changes (either deletions or amplifications) genome-wide.

- **Prognosis of malignant neoplasms.** Certain genetic alterations are associated with poor prognosis, and hence their detection allows stratification of patients for therapy. For example, amplification of the *NMYC* gene and deletions of 1p bode poorly for patients with neuroblastoma, and oligodendrogliomas in which the only genomic abnormality is the loss of chromosomes 1p and 19q respond well to therapy and are associated with long-term survival when compared to tumors with intact 1p and 19q but with EGF receptor amplification.
- **Detection of minimal residual disease.** After treatment of patients with leukemia or lymphoma, the presence of minimal disease or the onset of relapse can be monitored by PCR-based amplification of nucleic acid sequences unique to the malignant clone. For example, detection of *BCR-ABL* transcripts by PCR gives a measure of the residual leukemia cells in treated patients with CML. The prognostic importance of minimal residual disease has been established in acute leukemia and is being evaluated in other neoplasms.
- **Diagnosis of hereditary predisposition to cancer.** As discussed earlier, germline mutations in several tumor suppressor genes, including *BRCA1*, *BRCA2*, and the *RET* proto-oncogene, are associated with a high risk of developing specific cancers. Thus, detection of these mutated alleles may allow the patient and physician to devise an aggressive screening program, consider the option of prophylactic surgery, and counsel relatives, who may also be at risk. Such analysis usually requires detection of a specific mutation (e.g., *RET* gene) or sequencing of the entire gene. The latter is necessitated when several different cancer-associated mutations are known to exist. Although the detection of mutations in such cases is relatively straightforward, the ethical issues surrounding presymptomatic diagnosis are complex.
- **Guiding therapy with oncoprotein-directed drugs.** An increasing number of chemotherapeutic agents target oncoproteins that are only present in a subset of cancers of a particular type. Thus, the molecular identification of genetic lesions that produce these oncoproteins is essential for optimal treatment of patients. Current examples of genetic lesions that guide therapy and are frequently tested for in molecular diagnostic laboratories include the *PML-RARA* fusion gene in acute promyelocytic leukemia; the *BCR-ABL* fusion gene in chronic myelogenous leukemia and acute lymphoblastic leukemia; *ERBB1* (EGFR) mutations and *ALK* gene rearrangements in lung cancer; and *BRAF* mutations in melanoma.

Molecular Profiles of Tumors: The Future of Cancer Diagnostics

Until recently, molecular studies of tumors involved the analysis of individual genes. However, the past few years have seen the introduction of revolutionary technologies that can rapidly sequence an entire genome; assess epigenetic modifications genome-wide (the epigenome); quantify all of the RNAs expressed in a cell population (the transcriptome); measure many proteins simultaneously (the proteome); and take a snapshot of all of the cell's metabolites (the metabolome). Thus, we have entered the age of "omics!"

The most common method for large-scale analysis of RNA expression in use today in research laboratories is based on DNA microarrays, but newer methods that involve RNA sequencing have appeared that offer a more comprehensive and quantitative assessment of RNA expression. However, RNA is prone to degradation and is a more difficult analyte to work with than DNA in clinical practice. Furthermore, DNA sequencing is technically simpler that RNA sequencing, permitting the development of methods that rely on massively parallel sequencing (so-called next generation [NextGen] sequencing). The increases in DNA sequencing capacity and speed that such methods have enabled over the past decade have been breath taking, and are matched by equally remarkable decreases in cost. The first reasonably complete draft of the sequence of the human genome, released in 2003, took 12 years of work and cost about $2,700,000,000. At present, using NextGen sequencing, certain cancer centers are completing the process of whole genome sequencing for individual tumors in 28 days, which includes the time required for the extraordinarily complex task of assembling and analyzing the sequencing data. The cost of whole genome sequencing has now fallen under $3,000, and is continuing to drop; there is no doubt that the long promised $1,000 genome is now in sight.

These advances have enabled the systematic sequencing and cataloging of genomic alterations in various human cancers, much of it within a large consortium sponsored by the National Cancer Institute called The Cancer Genome Atlas (TCGA). The complexity of the genetic aberrations identified in these genome-wide studies has inspired informaticians to create new ways of displaying the data, such as circos plots (Fig. 7-50), which provide a snapshot of all of the genetic alterations that exist in a particular tumor.

The main impact of cancer genome sequencing to date has been in the area of research: identification of new mutations that underlie various cancers; description of the full panoply of genetic lesions that are found in individual cancers; and a greater appreciation of the genetic heterogeneity that exists in individual cancers from area to area. As noted, a few centers are piloting the use of whole genome sequencing to manage patients, but most efforts in the clinical realm are focused on developing sequencing methods that permit identification of therapeutically "actionable" genetic lesions in a timely fashion at a reasonable cost. Such approaches seem particularly applicable to