

Section E6.5 of the ACMG technical standards and guidelines: Chromosome studies for solid tumor abnormalities

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Disclaimer: This updated Section E6.5 has been incorporated into Section E6: Chromosome Studies For Acquired Abnormalities and supersedes the previous Section E6.5. Section E6 is part of Section E: Clinical Cytogenetics of the 2008 Edition (Revised February 2007) ACMG Standards and Guidelines for Clinical Genetics Laboratories.

These standards and guidelines are designed primarily as an educational resource for clinical laboratory geneticists to help them provide quality laboratory genetic services. Adherence to these standards and guidelines does not necessarily ensure a successful medical outcome. These standards and guidelines should not be considered inclusive of all proper procedures and tests or exclusive of other procedures and tests that are reasonably directed to obtaining the same results. In determining the propriety of any specific procedure or test, the clinical laboratory geneticists should apply their own professional judgment to the specific clinical circumstance presented by the individual patient or specimen. It may be prudent, however, to document in the laboratory record the rationale for any significant deviation from these standards and guidelines.

Purpose: Cytogenetic analysis of tumor tissue detects clonal abnormalities. The information obtained from these studies is utilized for diagnosis, prognosis, and patient management. **Methods:** The Working Group of the Laboratory Quality Assurance Committee of the American College of Medical Genetics provides these Standards and Guidelines for chromosome studies for solid tumors abnormalities as a resource for clinical cytogenetic laboratories. **Results:** The guidelines incorporate aspects of sample procurement, handling, processing, harvesting, analysis, quality control, and quality assurance. It is recommended that all pediatric solid tumors be studied by cytogenetic analysis when feasible due to the clinical and therapeutic implications of the genetic abnormalities. Cytogenetic analysis of certain adult solid tumors also provides information that impacts diagnosis and therapeutics. Molecular cytogenetic analysis or fluorescence in situ hybridization (FISH) may be a primary or secondary method of evaluation of the tumor tissue. FISH

can document a specific molecular event, e.g. gene rearrangement, provide a rapid result to aid in the differential diagnosis or planning of therapy, clarify chromosome anomalies, or assess gene amplification. **Conclusion:** Genetic analysis adds valuable information to the understanding of and therapeutic approach to solid tumors. Laboratories may use their professional judgment to make modifications or additions to these guidelines. *Genet Med* 2009;11(12):890–897.

Key Words: solid tumor, cancer, cytogenetics, chromosome, guidelines, fluorescence in situ hybridization

E6.5 CHROMOSOME STUDIES FOR SOLID TUMOR ABNORMALITIES

Cytogenetic analysis of tumor tissue is performed to detect and characterize chromosomal abnormalities for purposes of diagnosis, prognosis, and patient management.

E6.5.1 General considerations

E6.5.1 (a)

A patient with a solid tumor may have conventional or molecular cytogenetic fluorescence in situ hybridization (FISH) analysis of the tumor tissue at the time of biopsy or resection (to aid in differential diagnosis), at a time of disease recurrence (to confirm recurrence and to investigate disease progression), or to identify metastatic tumor or a tumor of uncertain origin. The solid tumor specimen may be accompanied by or followed by collection of other tissue samples (e.g., bone marrow and cerebrospinal fluid) for disease staging.

E6.5.1 (b)

The laboratory director and staff should be familiar with the chromosomal and molecular abnormalities associated with tumor types/subtypes and their clinical significance.^{1–5} Appendix

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A is a list of tumors divided into small round cell and non-small round cell tumor types. Appendix B includes common tumor chromosomal aberrations, known genes and FISH targets, and clinical significance as of the time of writing. Appendix B is modified from previous publications.^{6,7}

E6.5.1 (c)

The majority of pediatric tumors should be cytogenetically analyzed whenever sufficient fresh tissue is available because cytogenetic abnormalities are commonly disease or disease subtype specific and have prognostic significance. Cytogenetic analysis of selected adult tumors is indicated whenever such analysis may have diagnostic or prognostic value.

E6.5.1 (d)

Methods for processing of tumor material will be determined by the cytogenetic laboratory^{5,8} based on available clinical and pathologic findings. The cytogenetic laboratory should obtain as much information as possible about the suspected diagnosis and the tissue type at the time of sample receipt to choose the most appropriate tissue culture method(s). To the degree possible, the cytogenetic laboratory should communicate with the pathologist to gain information regarding tumor type by frozen section and/or permanent section analysis of the tissue.

E6.5.1 (e)

Molecular cytogenetic FISH analysis may be used as a primary or secondary method of evaluation of the tumor tissue. The availability of fresh tissue, the differential diagnosis, a need for rapid diagnostic information, and the type of information needed should be used to prioritize FISH relative to conventional cytogenetic analysis.

E6.5.1 (f)

Cytogenetic analysis results must be interpreted within the context of the pathologic and clinical findings.

E6.5.1 (g)

For quality assurance, the laboratory should monitor the number and types of tumors received, the percentage of tumors with abnormal results, the cell culture success rate, and the success rate for FISH studies.

E6.5.1 (h)

The presence or absence of specific abnormalities should be available to the physician as soon as is feasible to contribute to patient's plan of care.

E6.5.1.2 Specimen collection

E6.5.1.2 (a)

Solid tumor samples should be collected in a sterile manner. For conventional cytogenetic analysis, the tissue sample must be fresh. The sample selected for cytogenetic analysis should be pure tumor if possible, without necrosis. The sample must not be placed in fixative or frozen. (Specimens that will be evaluated solely by FISH analysis may be fixed, frozen, or paraffin embedded.)

E6.5.1.2 (b)

The laboratory should request a sample size of 0.5 to 1 cm³; if less tissue is available, the laboratory should accept as much as can be provided. If the sample size is very limited, e.g., fine needle aspirate or fine needle core biopsy, coverslip cultures are often successful. If, however, the sample size precludes cell

culture and conventional cytogenetic evaluation, the sample may be amenable to interphase FISH analysis using touch-preparations or paraffin-embedded tissue sections; see Section E6.5.2.1 (b) later.

E6.5.1.2 (c)

The tumor sample should be transported in culture medium to the cytogenetics laboratory as soon as possible for immediate processing.

E6.5.1.3 Specimen processing

E6.5.1.3 (a)

The cytogenetic laboratory should process the tumor sample as soon as it is received.

E6.5.1.3 (b)

The tumor sample should be inspected and details of the sample size, color, and attributes recorded.

E6.5.1.3 (c)

If obviously normal tissues are present, the tumor should be separated from nontumor tissue for processing.

E6.5.1.3 (d)

Disaggregation of tumor samples is needed for most tumor types. Mechanical and/or enzymatic methods may be used. If sufficient tumor material is submitted, both methods of disaggregation are recommended. For some tumor types, different growth characteristics can be seen with exposure to collagenase versus no exposure to collagenase. If sufficient material is available, cultures should be initiated with and without enzyme exposure.

E6.5.1.3 (e)

Culture methods, culture media, and culture conditions should be chosen to best support the type of tumor received. In general, tumors can be divided into small round cell tumors (SRCTs) and non-SRCT types (Appendix A). In general, SRCTs can be successfully grown in suspension and non-SRCTs are best grown with monolayer (flask or coverslip) culture methods. Most, but not all SRCTs will also grow in monolayer culture. If adequate tissue is obtained, both culture types should be initiated for SRCTs.

E6.5.1.3 (f)

The culture vessels used are chosen by the laboratory. Coverslips cultures may be used to successfully culture very small tumor samples. Duplicate cultures should be established whenever possible.

E6.5.1.3 (e)

Experience with tumor culture will provide the laboratory with information regarding optimal growth conditions for different tumor types. It can be helpful for the laboratory to maintain a database that documents how the different tumor types have grown and which culture and harvest conditions yield the abnormal clones. This database can then be searched for optimal processing and harvesting methods for any new tumor received in the laboratory.

E6.5.1.3 (f)

Frequent (daily) observation of cells in culture is needed to determine cell growth rate and optimal time to harvest. Tumor cells should be harvested as soon as possible on adequate

growth to capture early dividing tumor cells and prevent overgrowth by chromosomally normal cells.

E6.5.1.3 (g)

Conditions used for cell harvest will vary among tissue types, e.g., mitotic inhibitors used (colcemid, velban, ethidium bromide, etc.), their concentration, and exposure duration.

E6.5.1.4 Analytical methods

E6.5.1.4.1 (a)

Analysis of metaphase chromosomes should include cells with both good and poor chromosome morphology in attempting to identify an abnormal clone. Once identified, the clonal cells with the best chromosome morphology should be analyzed, karyotyped, or imaged to provide the most accurate breakpoint assignments.

E6.5.1.4 (b)

Clonal abnormalities should be documented in two independent cultures, if possible, to ensure that in vitro culture artifact is not mistakenly identified as a clinically significant abnormality.

E6.5.1.4 (c)

Cells that cannot be completely analyzed because of poor morphology should be scanned for obvious structurally abnormal chromosomes and abnormal chromosome counts.

E6.5.2.1

Analytical standards.

E6.5.2.1

Initial diagnostic studies.

E6.5.2.1 (a)

G-band analysis and documentation: Analyze 20 metaphase cells and/or a sufficient number of cells to characterize all abnormal clones and subclones.

For abnormal cells:

If only one abnormal clone: two karyotypes.

If more than one related abnormal clone: two karyotypes of the stemline and one of each sideline.

If unrelated clones: two karyotypes for each stemline and one for each associated pertinent sideline.

For normal cells:

If only normal cells: two karyotypes.

If normal and abnormal cells: one karyotype of a normal cell plus karyotypes for abnormal clone(s) as earlier.

E6.5.2.1 (b)

Molecular cytogenetic FISH analysis.

Tissue types

Sample types that may be used for FISH include (1) paraffin-embedded tissue sections, (2) touch preparations (TP), (3) cytospin preparations, (4) cultured or direct harvest tumor cells, (5) fixed cytogenetically prepared cells, or (6) fresh-frozen tumor tissues.

Paraffin-embedded tissue

- i. Before scoring a paraffin-embedded FISH slide, it is crucial that a pathologist review a hematoxylin and eo-

sin-stained slide and delineate the region of tumor cells that should be scored because it can be difficult to differentiate normal cells from malignant cells using only DAPI counterstain. The technologist should be clear, before scoring the slide, where the malignant cells of interest are located on the slide.

- ii. Formalin-fixed, paraffin-embedded tissue is acceptable for FISH analysis. Tissues preserved in B5 fixative or decalcified are usually not suitable for FISH.
- iii. Tumor sections cut 3 to 4 microns thick and mounted on a positively charged organosilane-coated (silanized) slides work well. Request several unstained sections and one hematoxylin and eosin stained slide from the submitting laboratory.

Touch preparations

A pathologist should make the TP or be involved in selecting the tissue for TP.

TP are helpful when tissue architecture is not crucial. TPs should be made by lightly touching the tumor piece to a glass slide without smearing. Air dry or fix in alcohol.

Cytospin preparations

Cytospin preparations are useful for concentration of samples with very low cellularity, e.g., cerebrospinal fluid.

Fixed cytogenetically prepared cells

Such preparations have multiple uses with both interphase and metaphase evaluations, including confirmation and clarification of suspected chromosome abnormalities or characterization of an apparently abnormal clone. Metaphase cell evaluation may help clarify specific chromosome rearrangements.

Fresh-frozen tumor tissues

Such tissues may be useful in sequential analysis of recurring tumors or in the evaluation of archived specimens.

Supplemental FISH analysis

As a supplemental test, FISH may be indicated to (1) document a specific molecular event, e.g., gene rearrangement that is diagnostic, (2) provide a rapid result to aid in the differential diagnosis or planning of therapy, or (3) to assess gene amplification. Characterization of the initial diagnostic FISH abnormality and signal pattern will provide a method for future assessment and monitoring of disease status.

Primary FISH analysis

FISH may be used as a primary method for tumor evaluation (1) when fresh tumor tissue is not available, (2) when rapid diagnostic information is needed to narrow the differential diagnosis, (3) to determine whether there is gene amplification for prognostic and/or therapeutic purposes, (4) when no metaphase cells are obtained by culture of tumor material, or (5) when conventional cytogenetic analysis yields a normal result.

Examples of such cases include, but are not limited to: SCRTs

- i. FISH with a probe for the *EWSRI* gene to identify tumors in the Ewing sarcoma family of tumors (EWS, pPNET, Askin tumor, and esthesioneuroblastoma) or other tumors with *EWSRI* gene rearrangement (e.g., clear cell sarcoma, desmoplastic SCRT, extraskeletal myxoid chondrosarcoma, and myxoid round cell liposarcoma).
- ii. FISH with a probe for the *FOXO1A* (*FKHR*) gene to identify alveolar-type rhabdomyosarcoma.

- iii. FISH with a probe for the *SS18 (SYT)* gene to identify synovial sarcoma.

Gene amplification

- i. FISH with a probe for the *MYCN* gene to assess the presence or absence of gene amplification in neuroblastoma.
- ii. FISH with a probe for the *ERBB2* gene to assess amplification in invasive breast cancer.

Differentiation of tumors with similar histopathology

- i. FISH with a probe for the *BCR* gene to detect monosomy 22 or deletion 22q. The *BCR* gene probe may be used as a surrogate for the *INI1* gene to differentiate atypical teratoid/rhabdoid tumors of infancy from medulloblastoma or extrarenal rhabdoid tumors from sarcomas.

Other applications of FISH will be determined on an individual tumor/patient basis to facilitate the diagnostic evaluation and monitoring of disease status.

Documentation

Documentation of FISH results should be in accordance with Sections E9 and E10 of these Standards and Guidelines for Clinical Genetics Laboratories.

E6.5.2.2

Follow-up studies

- i. May be indicated to assess recurrent disease or disease progression.
- ii. May be indicated to differentiate recurrence of a tumor from a new disease process.
- iii. Are indicated if the initial study failed.

E6.5.2.2 (a)

G-band analysis and documentation

- i. Analysis should include a minimum of 20 metaphase cells. Additional cells may be scored by G-banding or FISH for a specific abnormality identified at initial diagnosis.
- ii. Analysis should be performed with awareness of the possibility of a new clonal process, i.e., therapy-related malignancy.
- iii. FISH analysis may be recommended for diagnoses characterized by an abnormality for which FISH testing is available.

If both normal and abnormal cells or only abnormal cells:

One or two karyotypes from each abnormal clone with a minimum of two karyotypes.

One karyotype of a normal cell, if a normal karyotype was not documented in a previous study; otherwise, one normal metaphase spread.

If only normal cells: two karyotypes.

E6.5.3 Turnaround time

Turnaround time (TAT) should be appropriate for clinical utility. The cytogenetics laboratory may want to have a written policy describing how solid tumor cases are prioritized (with respect to each other and with respect to other sample types).

E6.5.3.1

Because of the multiplicity of tumor types and to variability of growth in culture, TATs will vary. However, the TAT for each

individual tumor should be as rapid as possible given such factors. Final results should be available within 28-calendar days.

E6.5.3.2

FISH analysis results should be available within 1 to 3 days for most tumors and 7 days for paraffin-embedded tissues.

E6.5.3.3

Preliminary verbal reports should ideally be given in 7 to 10 days, and the date of such results should be documented in the final report. The content of the preliminary report should be documented if it differs significantly from that of the final report.

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APPENDIX A

Selected solid tumors according to culture method

Tumors may be divided into SRCTs or non-small round cell tumors (NSRCTs) based on histopathology and whether the tumor is expected to grow in suspension (SRCTs) or as a monolayer culture (NSRCTs). Some tumors may grow with either method. Because the histopathology of a tumor is generally unknown at the time of receipt, this guide can help in deciding how to culture a tumor. If sufficient material is provided for a SRCT, culture with both methods; if only a small amount of tumor is received, it is safer to initiate the tumor culture as a monolayer particularly if coverslip culture is used. Suspension or direct harvest may provide material for FISH if culture growth fails.

Small round cell tumors

Suspension only tumors

Lymphoma
Plasmacytoma
Histiocytosis

Suspension and/or monolayer

Neuroblastoma
Retinoblastoma
Central primitive neuroectodermal tumor (PNET) or medulloblastoma
Ewing sarcoma, peripheral primitive neuroectodermal (pPNET)
Rhabdomyosarcoma
Osteosarcoma

APPENDIX A. Continued

Non-small round cell tumors

Brain tumors

- Ependymoma
- Glial tumors, glioblastoma, ganglioglioma
- Astrocytoma
- Oligodendroglioma
- Choroid plexus tumors
- Meningioma

Mesenchymal tumors or sarcomas or “spindle cell” tumors

- Hepatoblastoma, hepatocellular carcinoma
- Wilms tumor
- Malignant fibrous histiocytoma (MFH), fibrosarcoma
- Synovial sarcoma
- Clear cell sarcoma

- Desmoplastic small round cell tumor
- Liposarcoma, lipoma
- Hemangiosarcoma
- Leiomyosarcoma
- Mesothelioma

Germ cell tumors

- Teratoma
- Seminoma
- Embryonal carcinoma, yolk sac tumors

Epithelial tumors (carcinomas)

- Renal cell
- Breast
- GI
- Lung

APPENDIX B. Diagnostic/clinical significance of genetics of some solid tumors

Disease entity	Chromosomal aberration	Genes involved; FISH targets	Clinical significance
Genitourinary			
Renal			
Clear cell RCC	-3 or del(3p)	3p, <i>VHL</i> , other unknown gene	Characterize non-papillary RCC
	del(3p) with gain 5q	3p, 5q	Favorable prognosis
	del(3p) with loss 5q	3p, 5q	Metastasis, unfavorable
	-14/del(14q)	14, <i>IGH</i>	Unfavorable, shorter survival
Papillary RCC	+7, +17, -Y, 9p-	7, 17, Y, <i>CDKN2A</i>	Characterize adult papillary RCC
t(Xp11.2) RCC	t(X;1)(p11.2;q21.2)	<i>PRCC/TFE3</i>	Characterize pediatric papillary RCC
	t(X;17)(p11.2;q25)	<i>ASPLC1/TFE3</i>	Balanced translocation in RCC
	t(X;17)(p11.2;q23)	<i>CLTC/TFE3</i>	Characterize pediatric papillary RCC
	t(X;1)(p11.2;p34)	<i>PSF/TFE3</i>	Characterize pediatric papillary RCC
t(6;11) RCC	t(6;11)(p21;q12)	<i>ALPHA/TFEB</i>	Subset of RCC, children, young adult
Chromophobe RCC	Loss 1, 2, 6, 10, 13, 17, 21	Chromosome enumeration	Distinguish from oncocytoma
Oncocytoma	1p-, t(11q13)	1p, <i>CCND1</i>	Distinguish from chromophobe
Rhabdoid	-22/22q-	<i>SMARCB1</i> ; <i>BCR</i> as surrogate	Diagnostic
CMN	t(12;15)(p12;q25), +11, +17, +20	<i>ETV6/NTRK3</i>	Diagnostic
Bladder, papillary	del(9)(p21)	<i>CDKN2A</i>	Homozygous deletion higher grade, stage
	8p-	8p, unknown gene	Higher recurrence rate, progression
	+7, +17	<i>FGFR3, TP53, MYC</i>	Genetic instability
Wilms tumor	16q-, +1q, 1p-, -22, 17p-	1p, 1q, 16q, 17p, 22, <i>WT1</i>	Unfavorable histology; augmented chemotherapy if 1p-, 16q-
Prostate	+7q31, 8p22-, +8q24, 17p13-	7, <i>LPL, MYC, TP53</i>	High frequency in carcinoma
	17p13-, +8q24	<i>TP53, MYC</i>	Progression, higher Gleason score
	10q-	<i>PTEN</i>	High grade PIN or carcinoma

(Continued)

APPENDIX B. Continued

Disease entity	Chromosomal aberration	Genes involved FISH targets	Clinical significance
Gastrointestinal GIST	-14/14q-, -22/22q-	14, 22, <i>KIT</i> mutations	Distinguish from smooth muscle tumors Response to TKIs
Liver			
Hepatoblastoma	+20, +2, +8, t(1q12q21)	Chr 2, 8, 20, 1q	Distinguish from HCC, HMH
HMH	t(11;19)(q13;q13.4), t(19q13.4)	19q	Distinguish from hemangioma or malignant tumor
Salivary gland			
Pleomorphic adenoma	t(3;8)(p21;q12)	<i>PLAG1</i>	Diagnostic benign
Mucoepidermoid cancer	t(11;19)(q21;p13)	<i>MECT1/MAML2</i>	Diagnostic malignant
Warthin tumor	t(11;19)(q21;p13)	<i>MECT1/MAML2</i>	Benign tumor
Breast			
Invasive intraductal	dmin, hsr	<i>ERBB2</i> amp <i>ERRB2, TOP2A</i> co-amp	Worst prognosis, response to TKIs, Mab Co-amplification, better response to FEC
Secretory breast	t(12;15)(p13;q25)	<i>ETV6/NTRK3</i>	Favorable; distinguish from IDC
CNS			
Astrocytic tumors	+7, -10/10q- 9p21- 19q-	<i>EGFR, PTEN</i> <i>CDKN2A</i> 19q	Short survival, aggressive course Sensitive to antimetabolite therapy Long-term survival
Glioblastoma	+7, 10q-, 9p-	<i>PTEN, EGFR, CDKN2A</i>	Short survival assoc w/ 10q-/PTEN loss
Oligodendroglial tumors			
Anaplastic	1p-, 19q-, der(1;19)(q10;p10)	1p36, 19q13.3	Longer survival, sensitive to therapy
Mixed oligoastrocytoma	+7, -10/10q-, 15q-	<i>EGFR, PTEN</i>	High grade, progression
Oligoastrocytoma	19q- 1p-	19q13.3 1p36	Favorable outcome Favorable outcome
Ependymoma			
Spinal	+7, -22q, 14q-	7, 14, 22q, <i>NF2</i>	Distinguish subtype
Intracranial	+1q, 6q-, +7, 9p-	1q25, p16, <i>EGFR, CDKN2A</i>	Pediatric, high risk
MB	i(17q), 17p-, -10/10q-, +7	<i>MYCN, MYC,</i> <i>ERBB2</i>	Large cell/anaplastic morphology High risk
Supratentorial PNET	+1q, 16q-, 19p-	1q, 19p	Lacks i(17q), poor prognosis
AT/RT	-22 or del(22q11.23)	<i>SMARCB1; BCR</i> as surrogate	Distinguish from MB, PNET, CPC
Meningioma	-22 or del(22q11.2) 1p-, -14/14q-	22q, <i>NF2</i> 1p, <i>IGH</i>	Primary abnormality Increased risk of recurrence, anaplastic
Choroid plexus tumors			
Carcinoma (CPC)	Loss 2, 3, 4, 5, 6, 8, 10, 11, 13, 14, 15, 16, 17, 18, 19, 22	Chromosome enumeration	Distinguish from papilloma
Papilloma	Gain 7,8,9,12,14,15,17,18,19,20	Chromosome enumeration	Distinguish from carcinoma

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APPENDIX B. Continued

Disease entity	Chromosomal aberration	Genes involved FISH targets	Clinical significance
SRCTs			
Alveolar RMS	t(2;13)(q37;q14)	<i>PAX3/FOXO1A</i>	Older youth, poorer outcome
	t(1;13)(p36;q14)	<i>PAX7/FOXO1A</i>	Younger, extremity location
	t(X;2)(q13;q35); t(2;2)(q35;p23)	<i>PAX3/AFX, PAX3/NCOA1</i>	Variant translocations
		<i>IGF1R</i> amp	Disease progression
Embryonal RMS	Gain 2, 7, 8, 11, 12, 13, 20	Chromosome enumeration	Distinguish from alveolar subtype
	Loss 1p, 3p, 9q, 10q, 16q, 17p, 22	<i>IGF1R</i> amp	Gene amplification with anaplasia
Neuroblastoma	del(1p), del(11q) w/o <i>MYCN</i> amp	1p, 11q, <i>MYCN</i>	Unfavorable in Stages I, II, IVS
	del(1p), +17q, <i>MYCN</i> amp	1p, 17q, <i>MYCN</i>	Unfavorable all stages
	triploidy w/o above abn	Chromosome enumeration	Favorable
EWS/pPNET	t(11;22)(q24;q12) & variants	<i>FLI1/EWSR1</i>	Diagnostic, distinguish from other SRCTs
	t(21;22)(q22;q12)	<i>ERG/EWSR1</i>	
	del(9p), 17p ⁻ , der(1;16)(q10;p10)	<i>CDKN2A, TP53</i>	Unfavorable prognostic factor
DSRCT	t(11;22)(p13;q12)	<i>WT1/EWSR1</i>	Distinguish from other SRCTs
Clear cell sarcoma	t(12;22)(q13;q12)	<i>ATF1/EWSR1</i>	Absent in cutaneous MM
Retinoblastoma	del(13q14), gain 1q, 6p	<i>RBI, DEK, E2F3</i>	Hallmark of retinoblastoma
Lymphomas	Specific translocations		Distinguish from other SRCTs
Bone, soft tissue			
CFS/CMN	t(12;15)(p12;q25), +11, +17, +20	<i>ETV6/NTRK3</i>	Distinguish CFS from fibrosarcoma
Synovial sarcoma	t(X;18)(p11.2;q11.2)	<i>SSX1/SS18</i>	Biphasic-most <i>SSX1/SSX18</i> , unfavorable
		<i>SSX2/SS18</i>	Monophasic- <i>SSX1/SSX18</i> or <i>SSX2/SSX18</i>
Lipoma	t(3;12)(q27-28;q14-15), variants	<i>HMGA2/LPP</i>	Distinguish from LPS
Liposarcoma (LPS)			
Myxoid, round cell	t(12;16)(q13;p11)	<i>FUS/DDIT3</i>	Diagnostic for myxoid LPS
Myxoid	t(12;22)(q13;q12) variant	<i>DDIT3/EWSR1</i>	Variant of t(12;16)
Well differentiated	Rings, markers, dmin	<i>MDM2, CDK4</i> amplification	Distinguish from lipoma
Leiomyoma	t(12;14)(q15;q24)	<i>HMGA2</i>	Distinguish from leiomyosarcoma
ASPS	der(17)t(X;17)(p11.2;q25)	<i>ASPLC1/TFE3</i>	Unbalanced translocation specific for ASPS
EMC	t(9;22)(q22;q12)	<i>NR4A3/EWSR1</i>	Diagnostic, Tumor specific
	t(9;17)(q22;q11.2)	<i>NR4A3/TAF15</i>	Variant translocation
	t(9;15)(q22;q21)	<i>NR4A3/TCF12</i>	Variant translocation
	t(3;9)(p11;q22)	<i>TFG/NR4A3</i>	Variant translocation

(Continued)

APPENDIX B. Continued

Disease entity	Chromosomal aberration	Genes involved FISH targets	Clinical significance
Dermal tumors			
DFSP	der (22)t(17;22)(q22;q13.1) or r(22)t(17;22)	<i>COL1A1/PDGFB</i>	Distinguish from atypical DF, MFH Response to TKIs
GCF	t(17;22)(q22;q13.1)	<i>COL1A1/PDGFB</i>	Similar to DFSP
Bednar tumor	der (22)t(17;22)(q22;q13.1) or r(22)t(17;22)	<i>COL1A1/PDGFB</i>	Similar to DFSP
Hidradenoma	t(11;19)(q21;p13)	<i>MECT1/MAML2</i>	Same as Warthin tumor
Lung tumors			
NSCLC	3p ⁻ ,+7, <i>EGFR</i> high copy number or amplification	<i>EGFR</i>	Response to TKIs
Germ cell tumor			
Dysgerminoma, ovary	i(12p), 12p overrepresentation	12p	Distinguish from non-GCTs
TGCTs, seminoma, NS	i(12p), 12p amplification	12p	Most common aberration, invasive disease

RCC, renal cell carcinoma; CMN, congenital mesoblastic nephroma; PIN, prostate intraepithelial neoplasia; GIST, gastrointestinal stromal tumor; TKIs, tyrosine kinase inhibitors; HMH, hepatic mesenchymal hamartoma; HCC, hepatocellular carcinoma; Mab, monoclonal antibody; FEC, fluorouracil, epirubicin, cyclophosphamide; IDC, intraductal carcinoma; PNET, primitive neuroectodermal tumor; AT/R, T atypical teratoid/rhabdoid tumor; CPC, choroid plexus carcinoma; EWS, Ewing's sarcoma; pPNET, peripheral primitive neuroectodermal tumor; DSRCT, desmoplastic small round cell tumor; MM, malignant melanoma; CFS, congenital fibrosarcoma; ASPS, alveolar soft part sarcoma; EMC, extraskeletal myxoid chondrosarcoma; DFSP, dermatofibrosarcoma protuberans; DF, dermatofibroma; MFH, malignant fibrohistiocytoma; GCF, giant cell fibroblastoma; NSCLC, non-small cell lung cancer; TGCTs, testicular germ cell tumors; NS, nonseminoma; MB, medulloblastoma; SRCTs, small round cell tumors; RMS, rhabdomyosarcoma.